THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL:

CONTAINING

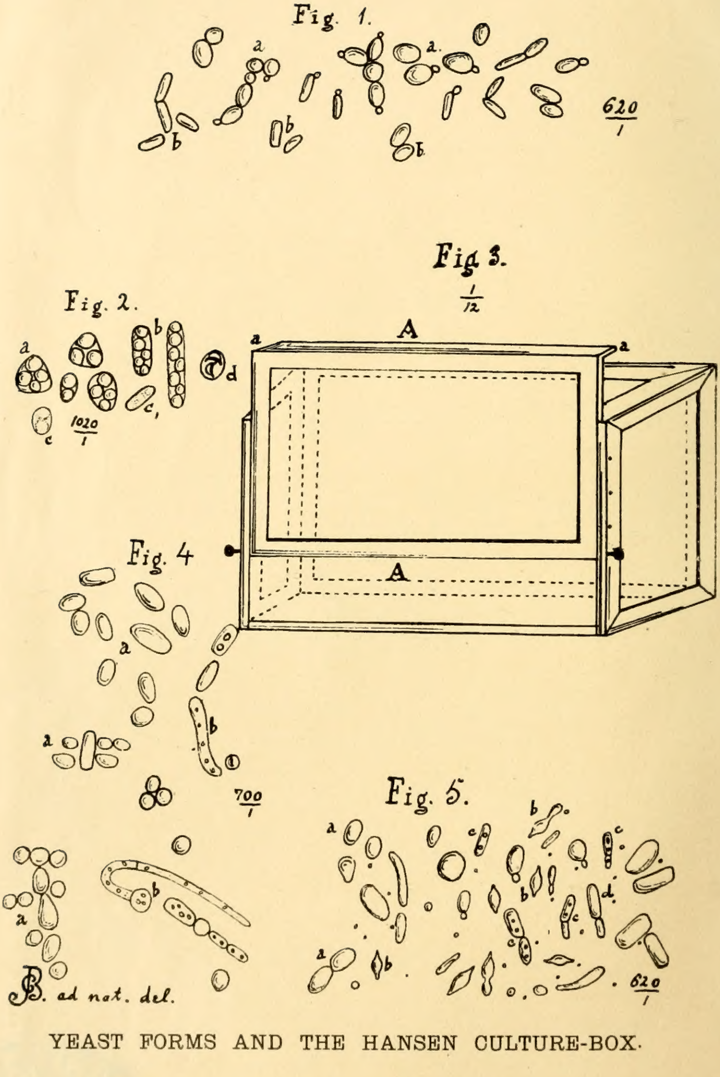
CONTRIBUTIONS TO BIOLOGY

VOLUME XV.

FOR

1894.

FOUNDED IN 1880, BY ROMYN HITCHCOCK, F. R. M. S.
PUBLISHED, SINCE 1887, BY CHAS. W. SMILEY,
WASHINGTON, D. C.



YEAST FORMS AND THE HANSEN CULTURE-BOX.

THE AMERICAN
MONTHLY
MICROSCOPICAL JOURNAL.

VOL. XV. JANUARY, 1894. NO. 1.

On the Study of Yeasts, With Descriptions of the Hansen Culture Box and of a New Infection Needle for the Study of Lower Organisms.

BY J. CHRISTIAN BAY,
Bacteriologist of the Iowa State Board of Health.
AMES, IOWA.

WITH FRONTISPIECE.

The study of the yeasts is one of the most interesting and instructive in botany. The material is easily obtained, and the student has ample opportunity to become acquainted with cell physiology. The yeasts are often studied in botanical and other biological laboratories, but a great many workers have not had the opportunity of becoming acquainted with the latest methods in this special branch, such as it has developed, during the last decennium, in the central laboratories for the physiology of fermentations, namely in the laboratories of Emil Chr. Hansen and Alf. Joergensen. The following notes will

EXPLANATION OF PLATE.

Fig. 1. a, cells of Saccharomyces cerevisiae I Hansen : b, cells of S. pastorianus I Hansen.

Fig. 2. Spore-forming cells : a, of S. cerevisiae I Hansen : b, of S. pastorianus I Hansen : c, of the same as in a ; formation of spores not yet completed, trace of spores : d, trace of spores in the same as in b.

Fig. 3. The Hansen Culture Box : AA, slide door with : aa, a cross-bar at the top.

Fig. 4. a, S. cerevisiae I Hansen : b, Monilia candida.

Fig. 5. a, Saccharomyces cerevisiae I Hansen : b, [Saccharomyces] apiculatus Reess : c, Mycoderma cerevisiae : d, S. pastorianus III Hansen.

serve as an introduction to the paper on the spore-forming yeasts which was published by the writer* some time ago.

The first question is how to obtain the proper material for investigation. Here, a mistake is often made; the so-called yeast-cakes manufactured and advertised as "manufactured of pure yeast" are by no means pure; everybody who knows a little of pure cultures and mycological work can see this. The yeast-cakes are very often made of yeast, (but by no means pure), a fair amount of starch, and a fair amount of bacteria is also found. It might be added that there are teachers who let their students work with this material, and who tell them that the starch-grains visible in the field of the microscope serve as a food for the yeast cells which break up the grains and form carbon dioxide and alcohol. The only place for obtaining good materials for study is the *brewery.*

A 200 c.c. bottle, sterilized, and wrapped in sterilized filter-paper, is taken to the cellar of the brewery where the fermentation takes place. It is unpacked and, by means of a sterilized string, lowered into one of the vessels. When it is nearly filled, it is drawn up under the surface of the fluid, and corked, before it is taken out in the air. It may be well to take two samples from the same vessel, one a few inches under the surface, and one at the bottom of the vessel. Two clean sticks will facilitate the latter, one fastened to the bottle, and the other one to the cork. Experience teaches the manipulation. Two such samples should be taken at the beginning, and two at the end of the fermentation. The flasks should be carefully packed and taken to the laboratory. If the preliminary examinations cannot be made at once, the flasks may be kept in an ice box for a few days.

*The spore-forming species of the genus *Saccharomyces.* (American Naturalist, XXVII, p. 685-696, August 1893).

LABORATORY STUDIES.

The first step in examining the yeast sample is the microscopical examination. The Leitz microscope, oc. 2, obj. 6 or the excellent Seibert lenses, oc. 0-3, obj. 3 and 6 give good and clear pictures; any microscope with good lenses, and about 600 diam., magnif., will prove satisfactory.

It is desirable to have 15-20 glass rods in a rectangular tin box. These can all be flame-sterilized at once; a glass with 1 per cent corrosive sublimate solution should be kept at the table for the rods out of use. The clean and sterilized tin box is to be covered with a thick glass plate. It should be remembered that the bottles must be well marked with numbers corresponding to the numbers in the note book.

The bottles must not be shaked very much when they are taken out of the ice box as the cork might come off and the contents be infected from the air. The cork should be lifted cautiously, and a glass rod dipped down to the bottom of the bottle. When it has been taken out again rapidly, the drop hanging at its end is placed on a slide; a cover is put over it, and we have now a sample ready for microscopic examination.

Before entering upon the various organisms which might come under observation, it ought to be remembered that drawings of everything seen should be put down in the note book.

The examination of a sample of yeast from the brewery will be illustrated by the following example.

I. Microscopic analysis. This is the first step.

The picture might be as in Fig. 1. It is evident that we have here two distinct forms of *cells*, *a* shows short rows of round or oval cells apparently in vigorous development, judging from their many buds; *b* shows oblong or rectangular cells, many of which are budding. The latter forms of cells are commonly called "pastorian

cells" or "cells of a pastorian form;" this term does *not* indicate, however, that they belong to any of the three species of the group *Saccharomyces pastorianus.*—We further remark that the round cells look more pellucid than the pastorian form does.

We should now reflect, what this might be. The question of the species is first to be settled, afterwards other questions arise.

We have before us *two forms of cells,* but still we might have only *one species.* We might also have two or more species; from the form of the cells we can draw no conclusions. It suggests itself that the round cells might belong to one of the cultivated forms *Sacch. cerevisiae I* Hans., or one of its varieties, and the pastorian cells to one of the wild forms; this cannot be decided but by means of a spore-cultivation.

II. The spore-cultivation necessitates the keeping of two thermostates, one at 25° C., the other at 15° C. Such thermostates are found in all laboratories, zoological or botanical. It is established that cultivated forms and other forms which generally occur from infection through the air (the so-called wild yeasts) form spores according to the following rules.

a. Conditions for spore-formation.

1. Defective nutriment.
2. Liberal aeration.
3. Temperature within the limits of optimum to both sides.

b. Relations of spore-formation to temperature.

On this occasion, attention is given only to the general analysis which answers the question *how to distinguish between cultivated and wild forms.* This is the most practical point, one most important in practical life where results have to be practically made use of in the brewery. The following table shows how to use the results when the analysis has been made.

	25° C in 40 h.	15° C in 40 h.	15° C in 72 h.
Cultivated yeast	——	0	0
Wild yeast	——	0	——

How is the spore-culture to be carried out?

When the sample for microscopic examination has been taken out, the bottle containing the yeast emulsion should be set apart, and the yeast allowed to settle at the bottom. After this, the fluid is almost completely emptied out, and only a few drops are left with the bottom layer of yeast. This may be done comparatively safely in the Hansen Box mentioned below. When the bottle has been flame-sterilized at the outside, it is placed in the box, corked.

The apparatus for spore-cultivation consists of a pair of small culture dishes* embracing a piece of gypsum, the dimensions of which are about $\frac{1}{6}$ of an inch smaller than those of the smallest of the dishes. Such apparatus should always be kept, steam-sterilized and wrapped in filter paper, in store. They are flame-sterilized on the outside, and put into the box with a wash-bottle of sterilized water, also flame-sterilized. In the box, the cover is taken off the dish with the gypsum, the yeast-bottle is shaken up, and one or two drops of the thick yeast-emulsion allowed to fall on the surface of the dry gypsum bloc. The cork is then replaced. After this, the gypsum is charged with sterilized water from the wash bottle.† The cover dish must, in accordance with rule 2 (see above) not fit closely, but air must be allowed to pass over the yeast layer. This is effected by a small piece of rubber, wood, etc., placed between the edges of the two dishes, and kept in the laboratory for that use in

*See Salomonsen's Bacteriol. Technology (Wood's Med. and Surg. Monogr. IV, 2, page 463.)

†Perfect soaking of the gypsum is recommendable, the water is, therefore, added twice, successively. When the bloc is soaked in the middle, this contion is fulfilled. Then, some water must stand around the gypsum (not *above* it), between this and the glass.

a small glass of dilute alcohol. According to a recent communication by Effront, squares of clay may be used instead of the gypsum. The form of the substratum is immaterial.

Cultivation in sterilized water in a test-tube, or on moistened filter-paper, will—ceteris paribus—result in a formation of spores in the yeast. For the determination of species, the method described above must be recommended, because it is one of the factors in Hansen's establishing of his species. It is still an open question whether the different ways of spore-cultivation affect the results in regard to the time, at constant temperatures, in which spores are formed.—Other points, concerning spore-cultivation of pure cultures, will be mentioned below.

REMARKS ON THE SPORES.—A table above indicates the results and how to use them. The spores are, in the cultivated forms very refractive compared with those of the wild forms*. Their size and number varies somewhat:

KINDS OF YEASTS.	Spores.		KINDS OF YEASTS.	Spores.	
	Dia.	No.		Dia.	No.
1. S. cerevisiæ I.	2,5–6	1–5	13. S. jœrgensenii Lasche.............. ...	1–2,5	2–4
2. S. pastorianus I....	1,5–5	1–10	14. S. conglomeratus reess	....	....
3. S. pastorianus II...	2–5	1–7	15. S. albicans Reess ..	....	1
4. S. pastorianus III.	2–4	1–10	16. S. Reessi David....	....	3
5. S. ellipsoideus I....	2–4	1–4	17. S. galacticola Perot. et Rib	....	2-4
6. S. ellipsoideus II...	2–5	1–4	18. S. I. of Will.........	1,5–5	1–5
7. S. marxianus.......	2–4	....	19. S. II. of Will.......	2–4	1-4
8. S. exiguus............	2–4	2–3	20. S. minor Engel.....	3	2–4
9. S membranæfaciens	....	....	21. S. ilicis Groenld....	....	......
10. S. ludwigii	....	1–8	22. S. aquifolii Groen.	....	....
11 S. anomalus .... [All of Hansen.]	....	2–4			
12. S. hansenii Zoph..	....	....			

*Wild forms is an expression now commonly used of the forms that are not cultivated. This term is intimately connected with the post-pasteurean view of these plants, and with the question of pure cultures and of pure yeast. Pure cultures, such as we know them to-day, were not known to Reess (1870)

Only Nos. 1–6, 11, 13, 18, 19, 20, 21, 22 occur in fermenting beer-wort; No. 14 is considered no distinct species. As a general rule, the protoplasm of the cultivated forms become aggregated on the gypsum; the spore-forming cells often form the so-called "partition-walls" or simply "walls" dividing the cell into 2–5 (generally only three) divisions, each of which contains a spore. This was never seen in the other forms among which *S. anomalus* partly figures as an exception by having half-formed spores (see Fig. 2 d). The wall consists of protoplasm pressed in between the spores, or there is a genuine wall which divides the mother-cell into two or more parts and changes it into a kind of sporangium or ascus.* Both cells and spores contain a nucleus.

THE HANSEN CULTURE BOX.

This box was mentioned by the writer at the Madison meeting of the A. A. A. S., in August, 1893. The box is rectangular (see Fig. 3), with panes in top and in the four sides. The bottom should be made of heavy, hard wood (hickory or pine), as it must not bend when moistened. In front a sliding door is fitted in tightly. This door can be placed in different positions, as the figure shows, allowing a bigger or smaller opening in front. About ten minutes before being used this box is washed carefully in and outside with a one per cent sol. of corrosive sublimate (Hg. Cl.), and closed. After a while the

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or to Pasteur (1876); "*pure yeast*, in the sense of Pasteur, means yeast free from *bacteria* and *moulds*, while, in the sense of Hansen, it means yeast free from infection of *wild forms*. Hansen's methods of pure cultures, and his system of "pure yeast" is now commanding the attention of physiologists as well of practical brewers, as it has been introduced in the leading breweries all the world over.

*The formation of spores here and in the *Phycomycetes* (*Saprolegnia*, *Mucor*, *etc.*) are different phenomena. Here, we have a free cell-formation the nature of which is partly unknown. (See DeBary, Comparative Morph. and Biol., p. 74, 268-9, 1887).

newly sterilized utensils are put into the box and arranged in a manner which experience will indicate. Then thə box is again closed. The hands of the worker are carefully washed, and the slide-door is raised so that it allows the hands and a part of the arms to conveniently work inside. A small acohol-lamp may be kept in the box for the final disinfection of needles, covers, slides, etc. But everything should be carefully sterilized before being put in.

This box is very convenient for a great deal of mycological work. It was first used by the famous physiologist at the Carlsberg Laboratory, Dr. Em. Chr. Hansen.

It should be added that a small cross-bar (a, a) prevents dust from falling into the box at the top of the sliding-door.

FURTHER STUDIES.

Pursuing the case before us we find that both of the two forms of cells which suggested the identification of cultivated yeast and one of the wild forms have formed spores after a time of 40 hours by 25.° The spore-cultures are taken out of the thermostat, and the cover lifted while a sample of the yeast layer at the top of the gypsum bloc is scraped off with a needle. This sample is then microscopically examined. In this case a diam. magn. of about 8–900 should be used. Seibert's oc. 3, obj. 3 always gives splendid pictures. Referring to Fig. 2, we find some cells with partition-walls and spores, indicating the presence of *S. cerevisiæ I.* Other cells of this yeast have no such walls (a); all these spores look, however, pale or "emptied." The other spore-bearing cells bear spores of a more refractive appearance, smaller than those mentioned above; these (b) evidently belong to one of the wild forms; the number of spores as well as the form of their mother-cell points towards one of the *S. pastorianus.* At c we find a spore-bearing cell of

S. cerevisiæ I with spores just being formed, and at d, we perceive the same feature of the spore-formation of the other form.

At present it is still an open question to which species the wild form belongs; this must be decided by a *pure culture.* The carrying out of a pure culture will be described in the second part of this paper.

We add two more examples of yeast-analysis before turning towards other questions.

Fig. 4 shows the microscopical picture of another sample. This time the mixture of different organisms is artificial. The analysis was given to the writer in Jorgensen's laboratory in Copenhagen. The figure was drawn from Seibert oc. 2, obj. 3, and shows two distinct types of yeast fungi cells, some round or oval, and some of a pastorian form. The clear, homogeneous protoplasm of the former indicates a culture yeast. At b we have a septated mycelium and cells with large oil drops; these might belong to a *Monilia* or *Chalara*, or similiar forms.

After 40 hours, some of the yeast cells had formed spores, the appearance of which (see above) show that *S. cerevisiae* is present. The cells that contained no spores had aggregated protoplasm. This shows that all of the yeast present is belonging to the above named species.

Much septated mycelium was found on the spore culture. A moist chamber culture from this, in a hanging drop of beer-wort, showed a typical *Monilia candida.*

Finally, we have another sample, the microscopic picture of which is demonstrated in Fig. 5. We find here (a) round or oval cells with homogenous protoplasm, like in a culture yeast*, (b) lemon shaped cells, some of which have a round bud at its end. This is undoubtedly Reess' *Saccharomyces apiculatus*† whose cells can always be

*Here, we are unable to say whether it is a variety of *S. cerev. I*, or not. The question of variation in this species is far-reaching, and has not yet been fully debated.

†This is one of Reess' species from 1870. Spores were never found in the

distinguished from those of the true yeasts.* At c we find some rectangular cells more refractive than those of a yeast, and with vacuoles; d shows cells of a pastorian form.

The spore-culture showed.

1. Spores in the round or oval cells by 25° after 40*h*. Spores also in the pastorian cells. The first are spores of a cultivated yeast, the latter not.

2. By 15° C.: After 40*h*. no spores, after 72*h*. spores in the pastorian cells, typical wild yeast spores.

An absolutely pure culture showed that this wild yeast was *Sacch. pastorianus III* Hansen.

The spore-culture was overgrown with a mass of rectangular cells of a low refractive power (grayish appearance). These belong to *Mycoderma cerevisiæ* Desmazieres,† and this species is not difficult to determine. On beer or wine exposed to the atmosphere, a gray, greasy looking zoogloea generally appears after a few days. The zoogloea soon becomes uneven at the surface. The cells are not easily separated, but aggregate in small lumps in the field of the microscope.

LITERATURE.

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cells, so it does not belong to the true *Saccharomyces*. The cells are always readily recognized on account of their typical shape.

*On the limitation of these, see Bay (Amer. Naturalist XXVI, p. 694-696, 1892).

†*Myc. cerev.* and *M. vini* cannot theoretically be separated from each other. Many text books state that these forms have spores; this was stated by J. de Seynes in 1868 and was repeated by others (see DeBary: Comp. Morph. and Biol., 1887, p. 268), though practical work shows that the *Mycoderma* does not have spores, and that, owing to the uncertainty of the species-question in 1868, De Seynes must have been working with true yeasts.

Flugge, Die Mikroorganismen, and Zopf, Die Pilze.

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AMES, IOWA, December 1, 1893.

(To be continued.)

A Mouldy Puzzle.

BY DR. ALFRED C. STOKES,

TRENTON, N. J.

I suppose that nobody cares how anybody else treats his apple, that most delightful of all the fruits, before he proceeds to eat it. Any one desiring to put a polish on the beautiful skin may do so, if he like, after the reported method of the street-venders, by the application of a little saliva and of much friction with the coat sleeve. But if you fail to open the apple before you eat it, you may swallow animal food with the vegetable: you will certainly devour a most beautiful fungus which you probably will never see unless you cut through the horny cells which contain the seeds. It was in this way that, after years possibly of eating fungi, I accidentally found a fine growth of this attractive mycelium, and equally fine microscopic object, about which I know absolutely nothing, but which I mention in the hope that the reader

will be able to puzzle it out; "and when found make a note of."

In the past winter as well as during the present, I have been seeing in the apples which I always keep in some quantity in my special "den," a cottony, white growth meandering over the inner surfaces of the horny cells which enclose the seeds, and clustered in little white patches on the surface of the seeds themselves. The object is in no special variety of apple, but in almost any kind, varying in abundance as also in its presence, in a way that of course I do not understand. But the growth, when it is present in any quantity, is conspicuous in elevated, fluffy ridges spreading in irregular lines over the cell walls, and gluing the seeds to those walls by its widening, or rather its thickening, increase. So far as I have been able to learn, the growth does not extend through the cells of the fruit, and therein is the puzzle.

How do those vegetable threads, for that is what they positively are—how do they get into the cells which enclose the seeds? Scrape off a little of the perfectly white and cottony object, mount it temporarily in a drop of water, and it immediately reveals itself as a cluster of those fungous threads called a mycelium, where each consists of distinct cells, colorless, except when scraped from the seeds, when the coloring matter from them will usually be found to have tinged the adherent mycelial cells, and having the surface attractively ornamented by minute, colorless, papilliform elevations. As far as I know these little papillæ are characteristic of this mycelium; I do not recollect that I have ever seen such appearances in any other growth of the kind, nor that I have ever read of them. The threads not rarely branch, and the colorless protoplasm within their cells is often divided into little trabeculæ formed by the interlacing of minute protoplasmic filaments which demand

a good, adjustable, one-fourth or one-fifth inch objective for their proper display. The appearance of the entire growth is exceedingly fine to the microscopist that is fond not only of "finding out things," but of seeing the beautiful wherever it may be. It is this which, as it seems to me, is one of the chief charms of the microscope, since the more microscopical study we give to a minute object, the more beautiful and impressive it becomes, while the more man's handiwork is magnified the greater become its imperfections, the rougher it appears and the more unattractive. It is not so with Nature's handiwork. The nearer we can approach to the intimate structure of that, the more perfect it becomes and the more beautiful. This internal apple-fungus is no exception to the rule.

But what is troubling me at present, and what I hope will also annoy the reader, is, How does that fungus get where it is?

There can certainly be no growth of the kind without a spore to start it. I have found no spores. There is even no trace of mycelial threads beneath the horny cells on the inner side of which they may be abundant. I have also failed to find any trace of the threads in the cells of the apple itself. Yet they must be there. That spores should get into those closed seed-capsules while the seeds are only ovules in the unripe ovary, is hardly thinkable. The surface of the mycelial threads is so beautifully roughened that they should be easily recognized among the cells of the fruit, if they were there, where I suppose they must be and where I hope some reader will find them. Beneath their points of attachment the horny wall seems to be minutely fractured, as though the threads had penetrated from the outside; yet in no case has the apple itself seemed to be diseased. It is possible, however, that these broken lines may be the result of the erosion of the growing mycelium.

After having seen these things and having learned so little about them, I received a communication from my correspondent, Dr. Henry Shimer of Mt. Carroll, Illinois, in which he says, that while examining the stigmas of apple blossoms he had been observing spores of a fungus, needle-shaped, about 1-250 inch long by 1-5000 inch wide, which seemed to be at work in a way not beneficial to the flower. It is possible that those spores may have some connection with the mycelium within the horny seed-vessels, this supposition being especially note-worthy in connection with what Dr. Shimer further reports, as follows :—

"I have examined several of the stigmata, now brownish, from the blossoms in which the pollen is dead from wet weather, and find much of the above fungus, the spores in places standing on end among the papillæ, densely packed together. I do not know, but possibly it may have a scientific name among the few who study these things, but for our purpose we may call it the apple-blossom mould, though it is a very different thing from the ordinary blue mould that is so common on our bread and pies, if kept too long in hot weather; that mould has roundish seed-spores, but in this the spores are long, slender, needle-like seeds; they are pale in the microscope, and contain a granular fluid or protoplasm.

Besides, I observe long filaments or roots of the mould, called mycelium, in the stigma and style, sometimes penetrating clear down to the apple. But I did not in any of my examinations find them in the ovary."

If the mycelium from those spores penetrate to the ovary, what is its form, and what is it doing, while the ovary is developing into the apple?

But the easiest thing in the known world is to ask questions. I hope some reader will be the "wise man" able to answer these and all that go before. Which, "when found, make a note of."

Measurement of Blood Corpuscles.

BY M. D. EWELL, M. D.

CHICAGO, ILL.

[Republished by request from the American Monthly Microscopical Journal for August 1885, pp. 150-1.]

For some time past I have been endeavoring, for my own satisfaction, to determine whether there is a constant average size of the human red blood corpuscles, with the view ultimately to determine whether it is possible, by means of micrometric measurements, to distinguish human blood from the blood of domestic animals.

In order that the results arrived at may be compared with those of other observers, I think it proper to state at the outset the methods and instruments employed.

The first requisite is obviously a correct standard of length, and the accurate determination of the value of the eye-piece micrometer used. This preliminary work has engaged much of my time and attention for several months past, and I have finally succeeded in obtaining two very accurate standards. The one of these which has been used as the standard of the measurements hereinafter given consists of lines ruled by Prof. W. A. Rogers, of Cambridge, Massachusetts (who is recognized as the highest authority upon questions of this sort), upon speculum metal at intervals of 1-2000 inch. The relative and absolute corrections of this standard have been determined by Prof. Rogers with very great accuracy, and the value of a division of the eye-piece micrometer described below was determined by taking an arithmetical mean of a long series of measurements of different intervals of 1-2000 inch, so as to eliminate as nearly as possible all errors of graduation and of measurement, and the value of one division of the micrometer was thus found to be .0000009925 or, approximately, 1-1000000 inch. The stand used, with mechanical stage and Abbe condenser

was made by Mr. Walter H. Bulloch, of this city, and is of the pattern styled by him the 'biological stand.'

The actual tube length was 8.91 inches from end of nose piece to upper end of draw-tube.

The cob-web eye-piece micrometer used was also made by Mr. Bulloch, the pitch of the screw being ½ millimeter, and the micrometer head being divided into 200 parts, which were read to 1-10 of a division.

The objective used was a homogeneous immersion 1-10 made by H. R. Spencer, of Geneva, N. Y., having a numerical aperture of 1.35, and it was used with a Bausch & Lomb achromatic amplifier, giving an amplification of about 1,500 diameters. The immersion fluid was Prof. Smith's new homogeneous immersion fluid, the composition of which he has not yet made public.

The blood was drawn from my finger, and a thin film spread with a needle upon the side of a cover-glass from .150 to .165 of a millimeter in thickness, and examined at once, a fresh sample being used upon each occasion. It was examined with central illumination, and always under as nearly the same conditions as possible. During the first four days of the examination, I took, night and morning, about one drachm of the elixir of calisaya, iron, and strychnia; during the rest of the time no drug was taken, and the conditions were nearly identical each evening. From 25 to 100 corpuscles were examined each evening, and I have tabulated the results, giving the smallest, largest, and average size, in millionths of an inch, of each 25 corpuscles; also the average of each 50, 75, 100, and 200 corpuscles. The corpuscles were measured, large and small, as they presented themselves in the field of the microscope, the only condition being that they should be approximately circular.

An examination of the above figures shows that the difference between the greatest and smallest averages of 25 corpuscles is .000028 or 1-35714 inch, a magnitude

that may be easily measured by any person having the requisite skill and apparatus.

The difference between the highest and lowest averages of 50 corpuscles is .000015 or 1-66666 inch, which approaches more nearly the limit of micrometric measurement, though probably not beyond it.

The difference between the highest and lowest averages of 75 corpuscles is .000012 or 1-83333 inch, which approximates the limit of micrometric measurement.

The difference between the highest and lowest averages of 100 corpuscles is .000009 or 1-111111 inch, which is within the limits of personal and instrumental error, according to the highest living authority upon this subject, who writes in substance, that it is easy to measure 1-50000 inch, but to be sure of 1-100000 inch is not possible.

The conclusion to be deduced from the above figures is obviously that, when a sufficient number of corpuscles are measured, there appears to be an average size which varies within very narrow limits, which may possibly be accounted for or at least is consistent with personal and instrumental errors; for though I have carried out the figures to the sixth decimal place, I have not the presumption to declare that the results can be relied upon farther than the fifth place, and have carried out the figures to the sixth only to insure accuracy in the fifth so far as possible. Another conclusion is, that granting for the moment that it is possible to identify blood by measurements of the red corpuscles, of which I am by no means satisfied, it is reckless in the last degree, if not criminal, to express an opinion upon the measurement of less than 100 corpuscles. To express an opinion upon the measurement of only 10 corpuscles, as I am informed has been done in this section within the last year or two, to take the most charitable view of the subject, betrays such culpable ignorance of a subject involving such mo-

nemtous consequences as ought forever to invalidate the testimony of one who should swear so recklessly. In a case involving the issue of life and death it would be better to measure several hundred corpuscles.

An examinaton of the unabridged table of measurements, from which the above summary is tabulated, discloses the further fact, that by selecting the corpuscles it would be possible for a dishonest observer to make the average much larger or smaller than that above given, without the possibility of detection ; a fact, the bearing of which upon the value of expert testimony upon this subject is so obvious as to need no comment.

It will be seen that I have not attempted to draw any inference as to the cause of the larger average size of the corpuscles first measured. Whether it was or not due to the drugs exhibited during the beginning of this work, is an interesting subject of inquiry, which must be reserved for future examination. I expect to continue these investigations, and at some future day will publish the result.

Sarcina Ventriculi in Blood Stains.

BY W. N. SHERMAN, M. D.

MERCED, CAL.

[Abstract from paper read before the American Microscopical Society.]

Goodsir was the first to describe in the vomit of some patients, peculiar groups of four cubical cells with rounded edges, and closely placed against one another. The flat surfaces of the groups are arranged in parallel layers and firmly connected by a gelatinous membrane. Klein states that they are occasionally found on boiled potatoes, egg albumen and gelatine exposed to the air, but that such sarcinæ are considerably smaller than the sarcina ventriculi found in the stomach. They may be successfully cultivated through many generations in pork or beef broth, or gelatine at ordinary temperatures. They

are of a greenish color, and often in large quantities have a yellowish tinge.

They are found in the stomach under normal conditions, but not so frequently as in disease, being most common in dilatation of this organ and in pyrosis, a form of dyspepsia characterized by acid fermentation and eructations of small portions of the contents of the stomach. They are also present in catarrhal conditions of this organ and chronic gastritis of long standing.

In pyrosis the fluid containing sarcina is often ejected from the mouth, and when existing in the fluids of the stomach they may be present in the saliva of the same subject (Budd).

The writer has observed this ejection of the fluids of the stomach, acid eructations, in a case of pyrosis existonly during periods of excessive indulgence in alcoholic liquors. Beale concludes that they are distinct from all other fungi, and in all cases coming under his observation the fluids containing them were distinctly acid.

Aitken asserts that pyrosis is frequent in Ireland and Scotland, and Linneus writes that one-half the inhabitants of Sweden are subject to it, a result of the large quantity of liquor drank in those countries.

The writer was summoned, as an expert, to examine the blood stains upon clothing of a man accused of murder. Those stains proved consistent with human blood, and not (as claimed) with the blood of a calf. There was also found imbeded in a clot the bulb and a small part of the shaft of a gray hair, the color of which corresponded with that of the murdered man. There were found also in these stains large numbers of the sarcina ventriculi, which the writer regards as tending to prove the identity of the blood.

It was proven that the murdered man was an habitual consumer of large quantities of acid wine, and had been for many years. The old man was murdered by having

his skull and face crushed with a mining hammer, and the circumstances (by a foot-print) showed that the lower part of the pants' leg, where stains were found, had been placed near to, or against, the face of the deceased, so that it is possible that the stain was mixed with the saliva or stomach secretions. Other portions of the murdered man's blood were not examined under the microscope, and the discovery of the sarcinæ was made accidentally and only in the blood stains on the pants of the accused.

The writer is not aware of the sarcinæ having been previously discovered in this manner and only reports it as of interest in a medico-legal way.

On the Development of the Continental Form of Microscope Stand.

By J. B. NIAS, M. D.

LONDON.

The first point of interest to note about the Continental stand is that it has maintained its form without substantial alterations for nearly fifty years—a proof, in general, that a design has been at the outset the creation of a practical man; and yet this stand presents several features which are open to criticism, and are in fact unfavorably criticized if taken as representing an optician's idea of what is suitable for a microscope; so that it became necessary to investigate the reasons for the steady preference shown for this stand on the Continent, in spite of such defects; and it soon appeared to me that they could only be explained as limitations introduced into the design at the bidding of some particular worker, such restrictions being submitted to by the optician, so that the stand may be regarded as one in which certain features desirable from the purely optical point of view have been deliberately suppressed in order

that other advantages may be gained. And as the partisans of this stand are chiefly found among the ranks of anatomists, it seemed reasonable to look for the original designer among them also, the result being that I have arrived at conclusions that appear novel and interesting, and worth communicating to others.

The Parisian optician Oberhaeuser is generally named as the inventor of this stand, but it is not easy to understand how an optician by himself could have arrived at a model so defective from the optician's point of view, and differing so widely from the other types of microscope in use at the time. Let us, however, suppose him to have worked under directions, and the matter becomes easy of explanation. Accordingly, upon investigation I met with repeated assertions, never effectively denied, on the part of a certain anatomist, that it was he who designed the original model of the Continental stand for his own particular use, and that it was made for him by the firm to which Oberhaeuser belonged, and, finally, that with his sanction it was patented by them, so as to give them for a certain period a monopoly of the manufacture—a fact which well explains its association with their names. I also find that this firm was permitted by the anatomist Dujardin to do the same thing with an achromatic condenser which he had invented, and, indeed, that they were actually the first opticians in France who conceived the idea of patenting inventions in connection with the microscope; but to substantiate this position requires the enumeration of a considerable number of details.

The anatomist to whom I have referred, Strauss-Durckheim by name, pupil of Cuvier, first addressed the scientific public with a work on the anatomy of the Coleoptera, in which the common cockchafer served as a type, which work being beautifully illustrated from his own dissections, aroused some curiosity, as he tells us

subsequently, on the part of naturalists as to the methods of investigation employed. To gratify this he undertook a second treatise on the methods of comparative anatomy, in which, among other instruments, are figured two microscopes which appear to me to present the earliest type, regard being had to date, of the Continental stand.

The work of dissecting small animals requiring the alternate use of the simple and the compound microscope with as little disturbance as possible of the preparation, together with its arrangement in a convenient form for manipulation, we find this point particularly studied in the design of these two stands: firstly, by the introduction of a rotary stage; secondly, by a limited height and a vertical position of the model. A precise account of his claims to this invention is to be found in a letter written by Strauss-Durckheim in 1850 to the optician Chevalier, which is printed in the life of the latter by his son; from it I quote the following paragraphs:

"Having formerly had to occupy myself a good deal with anatomical researches on very small animals with a microscope, which by a kind of chance, was of small dimensions, I did as other microscopists do, I tried to make the best of it by perfecting the mechanical part by several means which necessity suggested to me, and reflecting on all the inconvenience which I had encountered in my microscopical researches during my long experience, I finished by designing one of these instruments in which all difficulties were removed, and this was the microscope of which I published the description in my work on the art of dissecting which I mentioned above" (the Treatise on Comparative Anatomy, published it 1842, this being written in 1850).

He continues, "The first requisite of the microscope is to have in all about 3 dm. (= 12 inches) in height, so that the observer, comfortably seated at the table at

which he is working, and on which the microscope is placed, may have his hands on the stage of the latter where the object is which he is examining, whilst he looks into the eye-piece to see what he is doing while dissecting the object; which amounts to saying that the proportions which have appeared to me most suitable are those where the stage is raised about a decimetre (4 in.) above the table, and the entire tube of the microscope is only about 2 dm. (8 in.) long or a little more. The difficulty of arranging and suitably fixing the object which one examines being equally one of the greatest inconveniences in this kind of research, especially in the case of very small objects which a displacement of 1-10 mm. will cause to travel out of the field of the microscope, and which one ordinarily thus loses without being able to find them again, this inconvenience requires that such objects should be capable of being turned on themselves in every direction without displacing them from their situation in order to attack them from all sides. This advantage I have obtained by simply making the stage of the microscope movable on its centre. By this means, without touching the object itself, whether it be fixed or not, one can place it in all desired positions, and that without its receiving the slightest shock which might displace it. The observer requiring to have his hands firmly resting on this same stage while dissecting the object which he is studying, I have also found that the most suitable width for such a stage is that where it has about 1 dm. (4 in.) in breadth."

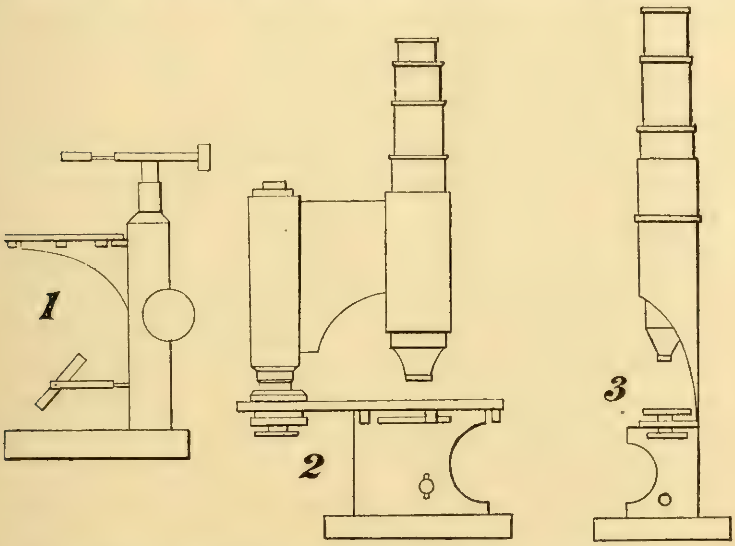
He goes on to state the requirements of such a microscope in a similar way at some length, but enough has been quoted to suggest that we meet here for the first time with a precise definition of what has become the standard dimensions of the Continental microscope stand.

Up to 1835, which appears to have been about the date

of the introduction of this model, there was no uniformity in the design of microscope stands, and it appears to me that this pattern succeeded in ousting all others by conforming in some degree, as described above, to the proportions of the human body, much as a spectacle frame is adapted to the face. One may be sure that for continuous and laborious use, such as falls to the lot of a professional worker, such an instrument as is least productive of muscular fatigue will be most fruitful in results. For it is evident that a man sitting at a table in the attitude of work will have his eyes naturally situated about 14 in. above the table, and about 6 in. from its edge, so that his arms, as they rest upon the table before him, will bring his hands close together in front, the directions of his eyes falling upon them without constraint, at an angle of about 15° from the vertical. A stand which falls into place within these dimensions will save fatigue—an advantage for which much may be sacrificed in many kinds of work. With a weighted foot, if produced slightly backwards, sufficient stability may be gained for such a small degree of inclination, or the vertical position will not be found extremely irksome, at least to the professional worker. The breadth of the hand regulates the height of the stage and its breadth: and the remainder of the space between the eyes and the table is all that is available for the length of the tube. Whatever opticians may say, those who have to economize labor will be found generally to prefer such a type of instrument; and the question arises whether it is more probable that such a design should have originated in the mind of an optician, or in that of a user of the microscope. I incline to the latter opinion, and may interpose the query whether any other reason can be given for fixing the tube-length for which Continental objectives are corrected at 150 mm. or thereabouts.

In fig. 1 the first of the instruments described by

Strauss-Durckheim is represented in outline. It is merely a dissecting microscope of common type with the important addition that the stage, which is carried by a movable bracket, is made to rotate, and is provided at its edge with several sockets, of which three are shown, for what we now term stage forceps, by which the various parts of the object under dissection are drawn asunder and fixed, while by rotation of the stage the preparation is turned into convenient positions for dissection. I do not find the right of Strauss-Durckheim



to be regarded as the inventor of this rotating type of stage anywhere disputed, and in the letter of Chevalier quoted the writer makes the explicit statement that the first microscope of the kind was constructed for him by the optician Cauchoix before 1824, four years before the publication of his work on the Coleoptera.

In fig. 2, the compound microscope is represented, and as being much more novel in form, it deserves a detailed examination. The foot is round and weighted with lead, and carries a cylinder or drum of brass with an aperture

at one side for the admission of light to the mirror, which, swung between pivots of which the ends project through the sides of the drum, is focused vertically by a rack and pinion of which the milled head is seen behind. At the top of the drum is a slit which gives passage to the edge of a circular diaphragm, and the stage, like that of the simple microscope, is round and fitted in the same way for numerous stage forceps; but, inasmuch, as its rotation with these in place would bring their projecting ends against the pillar of the microscope, the direct connection of the latter with the foot is severed, and it is inserted instead on a projecting lip of the stage upon which it is centered by being fitted loosely in a hole, where it is clamped in concentricity with the diaphragm by the clamping screw seen underneath. It is expressly admitted by Strauss-Durckheim that the working out of this feature was effected by M. Trecourt, Oberhaeuser's senior partner; and here we may ask for the original source of such a model. The inventor having stated that he originally worked "with a microscope which, by kind of chance, was of small dimensions," there was only one pattern extant at the time, which corresponds to this description and could have served as a basis for the design. This is the so-called drum microscope, shown in fig. 3, which is now only used for toys, but was adopted by the optician Fraunhofer, of Munich, as a regular pattern about 1815, and was certainly manufactured in Paris by Chevalier, Lerebours, Trecourt, and perhaps others from 1830 onwards; and it is not difficult to see how the combination of the stand with a rotating stage would result in the design of fig. 2. Returning to this instrument it will be noticed that the pillar which carries the body is enveloped in two outer tubes or sleeves, the inner of which, by means of a micrometer screw and spring, slides up and down to afford the now familiar form of fine-adjustment, whose milled head is seen below

the clamping screw which its stem perforates ;. while the outer is intended to allow the body to be swung to one side through an angle of 90,° when a simple lens mounted on another stand is to be brought over the dissection—a necessity only likely to be foreseen by a anatomist. The tube of the microscope itself is so evidently adapted from fig. 3 as to need no observation, and the only novelty is that it is double within, so as to permit of the use of a second objective above the first, as an erector, an original feature for which Strauss-Durckheim can also claim priority.

(*To be concluded in February.*)

The Bacillariaceæ or Diatomaceæ.

By ARTHUR M. EDWARDS, M. D.

NEWARK, N. J.

As it is intended, if the subscribers are enough in number, to publish a treatise or book on the Diatomaceæ, and as it is proposed to use for the title a new reading of the name, namely the Bacillariaceæ, it will be well herein as well as in the future to explain what is meant by the term used to designate this wonderful and beautiful group of organisms.

Availing himself of the writings of the late Rev. Eugene O'Meara, in 1872, in the Quarterly Journal of Microscopical Science in his "Recent Researches in the Diatomaceæ" when reviewing Dr. E. Pfitzer's "Untersuchungen uber Bau und Entwicklungen der Bacillariaceen (Diatomaceen)" he pens the following.

The name Diatomaceæ has been used by nearly all the more recent authors to designate the group. Rabenhorst, in his more recent work "Flora Europaea algarum aquae dulcis et submarinæ," 1864-1868, has adopted the name Diatomophyceæ for the group. But in his former treatise Die Susswasser Diatomaceen 1853 he used that of

Diatomaceæ. In this he has been followed by Grunow, Heiberg, Schuman, Cleve, and Suringar. Dr. Pfitzer, however, maintains that the name Bacillariaceæ should be substituted, the genus Bacillaria having been established by Sinclair in 1788; whereas the genus Diatoma was established by De Candolle in 1805; and some of the older writers on the subject have used this designation. It may be deemed inconvenient now to abandon the name of the group which has been so generally adopted by recent writers, but, on technical grounds, Dr. Pfitzer's view is undoubtedly correct and we chose the right on technical grounds to designate them the Bacillariaceæ, and they shall because they must be so designated.

Now what is known as a group or family of Bacillariaceæ? Let us go back to Aristotle as the father of Natural History. He classified animals, for animals were capable of classification. Vegetables, rocks and earth were of small moment then. They did not think, move, and have their being it was thought. He first attempted the scientific division of the animal world. His outlines are rude of course but this acute observer admitted but two great sections of animals; The highest creatures possessing blood (i. e. red blood), which corresponds to the Vertebrata of modern authors, and animals provided with a colorless fluid instead of blood, and corresponding to the Invertebrata of more recent zoologists. Pass we now to consider the naturalists as they must be considered and John Ray or Wray (as he wrote his name till 1670), was born in 1628, and he is remarkable as being the first to define species although he did classify animals and plants. His definition of species rested until it was overthrown by the researches of Darwin in 1857. In 1766, came Linnæus and he classified animals and plants with a positiveness not thought of before. He defined species as something positive and readily to be distinguished and ranked families also as a group

of things below kingdoms of which plants were. Thus plants were a kingdom and animals were a kingdom and rocks were a kingdom. Thus as plants or vegetables were a kingdom, they were divided into flowerless and flowering plants, and flowerless plants were divided into other divisions until we come to algæ or water plants without flowers, and these are divided into families of which one covers the plants in question, the Bacillariaceæ. These in their turn are divided into genera and the genera into species. So the Bacillariaceæ are a family of algæ or water plants without evident flowers. That is to say that was the place they bore in the plan of nature but now they are Protista or organisms which are midway between vegetables and animals. For Hackel, a German Naturalist, must be claimed the credit for placing them there and I am disposed to place them there also.

They mostly have shells composed of Siliceous rock-like earth so that they can be preserved almost indefinitely and their shells are known as Diatoms and are found in the carboniferous and triassic periods. But more commonly in the Tertiary and constitute the Infusorial Earths of Richmond, Va., and Santa Monica, Cal., besides many other points on the surface of the globe. Being very small and finely marked they came to be used as test objects for microscopic lenses so that one of them, *Pleurosigma angulata* is widely known. Their natural history and how they occur and their place in geology along with how to get and preserve them will be told hereafter.

EDITORIAL.

Stone Under the Microscope.—It is often held that the best method of determining the probable durability of a building stone is to study its surface, or thin transparent slices, under a microscope. This method of study in recent years has

been most fruitful in developing interesting and valuable knowledge of a scientific and truly practical character. An examination of a section by means of the microscope will show not merely the various substances which compose it, but also the method according to which they are arranged, and by which they are attached to one another. For example, pyrites is considered to be the enemy of the quarryman and constructor, since it decomposes with ease and stains and discolors the rock. Pyrites in sharp, well-defined crystals sometimes decomposes with great difficulty. If a crystal or grain of pyrites is embedded in soft, porous, light-colored sandstones, its presence will certainly demonstrate itself by the black spot which will form about it in the porous stone, and will permanently disfigure and mar its beauty. If the same grain of pyrites is situated in or near very hard, compact, non-absorbent stone, the constituent minerals of which are not rifted or cracked, this grain of pyrites may decompose and the products be washed away, leaving the stone untarnished.

MICROSCOPICAL APPARATUS.

Indestructible Clay Wick.—A lamp wick made entirely of clay giving 25 per cent more light than cotton wick; made capillary by incorporating with the clay while in a plastic state, filaments of unspun vegetable fibre which are burned out in the process of baking the clay.

The object is to provide an indestructible wick which shall possess all the advantageous qualities of an ordinary cotton or fibre wick; but which shall in addition last an indefinite time without renewal or necessity of trimming or care.

When the clay is baked the vegetable fibre is burned out, leaving capillary tubes running longitudinally through the wick through which the oil from the lamp will be raised to the flame by capillary attraction. Owing to the perfect combustion of the wick, the flame is perfectly white in character, devoid of odor and smokeless. It is found through a practical test, that the oil is volatilized by the use of this wick and the vapor is consumed thus giving the above results.

Unspun vegetable fibre, owing to its fineness of thread is superior to all other filaments, the manufactured being too

coarse to produce proper capillary tubes as proven by actual tests. These wicks can be made in any desired shape or size, either by hand or the use of machinery adapted for the purpose. They have been used by Dr. Ephraim Cutter and Dr. John A. Cutter in microscopical work, clinical and laboratory, for months; and with objectives including the one-sveenty-fifth inch of Tolles. They state that the light is the best afforded them for such work. Since there is no combustion of cotton as in a cotton wick the wick does not clog hence needs no trimming.

MICROSCOPICAL MANIPULATION.

Moisture on the Cover Glass.—The cause of a deposit of moisture on the under side of the cover-glass must be sealing up before the object or the base on which it lies is thoroughly dry, or perhaps through the ring not being cemented properly on the glass slip, and so allowing the medium in which the finishing cement is dissolved to get through into the cavity in the cell in which the object lies, and condensing on the cover. The cure: With a sharp knife scrape off the ring of cement which lies on the surface of the cover-glass, also slightly down the side, about the thickness of the same; then warm the cover-glass slightly over a small spirit lamp, moving it about to prevent cracking it, and it should be then easily removable with the knife without injury either to the glass or object; then, before replacing, take care that the object is thoroughly dry. If the ring springs off instead of the cover-glass only, dry thoroughly; then put just sufficient cement on the underside of it to attach it to the glass, and let this dry thoroughly before finishing off.

BACTERIOLOGY.

The Typhus Bacillus.—Dr Fraenckel of Berlin, announces that he has discovered a typhus bacillus; and that by using it in vaccination, he has produced a rapid, benign course of the fever. Dr. Rumpf has cultivated an anti-fever bacillus which, he says, will cure typhus in eight days.

To Detect Influenza Bacilli in Blood.—Spread the blood upon cover glass slips. Dry thoroughly. Put in absolute alcohol 5 to 10 min. Stain as follows:

Take 40 grammes of aqueous solution of methylene blue; 20 grammes of ½ per cent solution of eosin dissolved in 70 per cent alcohol; 40 grammes distilled water. Place the cover glasses in this solution and heat in an incubating oven at 37°C from three to six hours. Then wash in water, dry and mount in balsam. The eosin stains the red blood corpuscles red and the leucocytes blue. The bacillus is seen in these as short rods often resembling diplococcus.

MICROSCOPICAL SOCIETIES.

Birmingham Microscopists' and Naturalists' Union.

September 18.—A series of microscopic fungi and one of cup corals cut and polished in two directions were shown. Also a collection of *Limnæa peregra* showing various stages of malformation. Mr. Nelson believed that some of these forms were due to a sudden influx of food which caused the shells to enlarge very quickly; afterwards when there is a scarcity of food the shell is too large and septa are formed to cut off inconvenient corners.

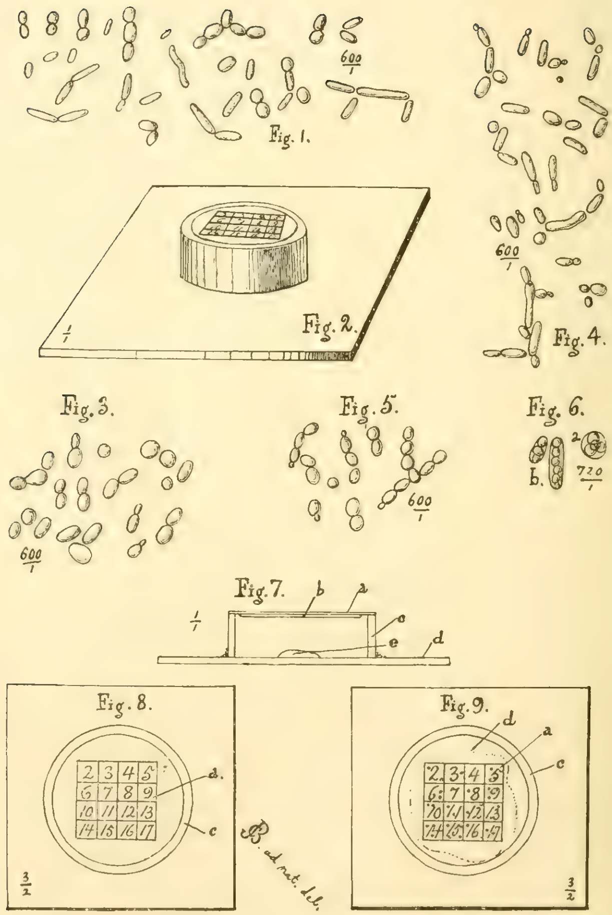
NEW PUBLICATIONS.

Illustated Guide to British Mosses. By H. G. Jameson. pp. 83, 59 figures.

The introduction treats of (1) the moss plant in general, (2) the stem and its appendages, (3) the leaves, (4) the capsule. (5) the spores, (6) the inflorescence. Then follow keys to genera and species and directions for examination of specimens. This is the only complete key to species that has been published. The book costs $2.00 and is to be had from the author whose address is Eastbourne, England.

Report on the Microscopic Examination of Blood from a Patient Suffering from Splenic myclogenous Leukæmia. By Frank S. Aby. Reprint from Bulletin of State University of Iowa, pp. 311-321.

We have not space to notice this important paper but recommend all specialists studying the blood to send to Prof. Aby at Iowa City for a copy. He has made some very careful counts and measurement of corpuscles and leucocytes.



THE CULTIVATION OF YEAST.

THE AMERICAN MONTHLY MICROSCOPICAL JOURNAL.

VOL. XV. FEBRUARY, 1894. NO. 2.

On the Study of Yeasts, With Descriptions of the Hansen Culture Box and of a New Infection Needle for the Study of Lower Cryptogames.

BY J. CHRISTIAN BAY,

Bacteriologist of the Iowa State Board of Health.

DES MOINES, IOWA.

(*Continued from page 11.*)

In the first part of this paper we have outlined the means of obtaining endospores in yeast. The budding is the main form of propagation ; only in three species of those known at present, spores occur in the fermenting fluid, viz. : *S. membranaefaciens*, *S. ludwigii* and *S. anomalus*. We came so far that we were able to distinguish between a cultivated and a wild form ; this is all that is required for the common yeast analysis in breweries. The number of "wild" cells may be ascertained by examining the spore culture by means of a hæmatimeter ;* it is impossible to obtain useful results

EXPLANATION OF PLATE.

Fig. 1. Microscopic picture of yeast sample submitted to pure cultivation.

Fig. 2. Boettcher's (model of a) moist chamber, adapted to the Hansen method of absolute pure cultivation (see text).

Fig. 3. S. cerevisiæ I (var. easily forming spores).

Fig. 4. S. Pastorianus III.

Fig. 5. S. cerevisiæ I (var. not easily forming spores).

Fig. 6. a. Spores of 3.
b, same of 2.

Fig. 7. The moist chamber, vertical section; a, cover; b, gelatine layer; c, glass ring; d, slide; e, drop of water.

Fig. 8. The same seen from above before inoculation.

Fig. 9. The same after inculation; 27 mature colonies seen through the slide. The two colonies in plots 6 and 10 are too close together, and should not be used in the preparation of the flask cultures. The outlines of the gelatine layer are indicated (d). a, slide of the chamber; c, glass ring in Figs. 8 and 9.

*See Jenaische Gesellsch. f. Medicin und Naturwiss, 1878, p. 1–18.

by merely counting the number of pastorian and oval cells with a pale, "wild" appearance. The percentage of wild forms among the cultivated ones can be ascertained successfully only from the spore-cultures where the spores allow comparison.

When the yeast is taken from the brewery directly into the laboratory and examined, it is vigorous and in good development; therefore, it can be cultivated for spores at once. If we have, on the other hand, a pure culture, this must be "freshened up" in beer-wort for 24 hours before it is transferred to the gypsum. This is done in the way that a trace from the original culture is brought into a new flask with sterilized beer-wort and kept by 25°C for 24 h.

The Freudenreich flask is the best for common use (see Salomonsen's Bact. Technol., l. c. p. 456, Fig. 10, to the right), but the Chamberland flask or a common test tube also work well. In extensive and important studies, the Pasteur flask (latest modification) is indispensable but more difficult to manipulate than other flasks. Among the many media prescribed and used none is better or more natural than beer-wort (gehopfte Wuerze in German breweries), the wort generally to be had in breweries is too concentrated, and sterile water should be added (from ½ to 1 vol. of water to 1 vol. of the wort). It must be sterilized and carefully filtered until it is absolutely clear and then poured into the small flasks or tubes.* To these a little sterile water is added and then they are again sterilized as other media. Before they are put aside they are examined every day for three or four days. If any vegetation appears they must be re-heated, water being added in order to prevent excessive evaporation. The laboratory must be kept closed and no windows opened while the pure cultures are made.

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*Everything must be sterilized before being used. See Salomonsen, l. c. p. 440.

THE PURE CULTURE.

Pure cultures of yeasts begin with a microscopical examination of the sample in order that the results may be somewhat estimated. We shall again illustrate by an example.

The microscopical picture of a yeast sample is demonstrated by Fig. 1. Three forms of cells are found, (a) round and oval, isolated culture-yeast cells, (b) round and oval cells in small rows, (c) pastorian cells, isolated or in rows.

A few drops from the yeast flask are deposited in a Chamberland or Freudenreich flask half filled with sterilized water. This is shaked violently for a couple of minutes, and the contents have then a milk-and-water like appearance. From this flask a drop is again transferred into sterile water, stirred, and from this second flask a third and similar transfer is made. These water flasks are previously marked and numbered. The two last flasks remain in the culture box; the whole process should be carried out here. The water in the last flask contains a number of cells pr. cc. very insignificant when compared with the original emulsion. If a drop from this is examined microscopically, only one or two cells must be observed within the field of the microscope. If there are more, another transfer is made, a suitable amount of the emulsion in flask No. 3 being transferred in order to make sure the result. When the transfer has been made, the flasks should always be stirred.

While this takes place, a 10 per cent beer-wort gelatine is ready for use in a water bath by 40°C. which tempature keeps it liquified. When ready for use it is cooled down to 30°C. and taken into the Hansen box where the following apparatus is waiting:

1. The yeast emulsion containing the proper amount of cells.

2. A big moist chamber. (See Fig. 2).

3. A glass of pure vaseline, paraffine and a brush.
4. Infection needles.
5. A big slide (2x2 inch).
6. A tin tray (4x3 inch).
7. A forceps.
8. A bell-glass.

All properly sterilized. The moist chamber is about one inch in diameter, and the cover is fastened to the upper end of the glass ring (see Fig. 2, a). The cover is further divided into 16 squares and the numbers 2-17 engraved in these. Such moist chambers are made by Mr. Jacob, Hauser Plads, Copenhagen. The beer-wort gelatine is thus made: 100 grammes of beer-wort is heated with 10 gr. of gelatine, and, when near to the boiling point, poured into a hot water funnel (see Salomonsen, l. c. p. 467), and filtered. The flame which heats the funnel is kept low and the fluid not allowed to boil. When filtration is over, the gelatine is again heated, poured into sterilized flasks and sterilized in the usual way.

Now the yeast emulsion is again stirred, and a drop transferred into the (30°C.) gelatine; the gelatine flask is gently stirred. The big moist chamber is laid down on the tray, cover downwards, and some vaseline applied to the edge of the glass ring pointing upwards. Then a small amount of the gelatine into which the yeast emulsion was transferred, is taken out with a glass rod and placed at the under surface of the cover facing upwards of the moist chamber, where it is carefully spread out in a thin layer. It is left in this position to become solid, being covered in the meantime with the bell-glass. When the gelatine is solid, the chamber is inverted and the ring pressed down gently upon the big slide. The vaseline prevents the air from passing in and out. A drop of water must be placed on the slide before the ring is inverted and laid down upon it, in order to keep the

chamber moist. The chamber is now ready for immediate microscropic examination under a low power, e.g. 300 diam. The object is to find and mark such cells as are laying in the gelatine separate from other cells. Consequently every plot on the cover is carefully searched. Beginners should take care to focus down first, so that the cover is in focus, and then go slowly further until the gelatine layer covering the under surface of the cover is reached. Here isolated cells are to be searched for. It is well to draw a big square in the note book, divide it into sixteen squares and draw the figures of the numbers just as they appear in the field of the microscope. In the respective places in the small squares in the note book, the isolated cells are then marked as they appear in the gelatine layer perpendicularly under the squares engraved on the cover glass of the chamber. Whenever a cell has been noticed, its surroundings in the gelatine are carefully searched and the following points strictly followed :

(a). The whole gelatine layer must be searched, not only the part which is in focus. Healthy looking cells are marked in the note book with an o.

(b). Two cells situated close together, at the side or above each other, are noted ∞ or 8 according to their position, but although their growth is watched, they are not used afterwards.

(c). Two cells within the same limits of the field are not used, but must be marked o-o.

(d). Dead cells or apparently dead cells are marked x. They are also watched in the course of the development.

When every field or plot has been examined, or when 25-30 cells conveniently situated have been marked, the moist chamber is placed beneath a bell glass and put aside. It must not be placed by 25°C. in the thermostat, but kept below 20°C., as the gelatine becomes liquified by a temperature a little above 22°C. The marked cells

are watched every two or three hours, and as the cells propagate and small colonies appear in the gelatine after a couple of days, the surroundings of the colonies should be watched, and the aspects of the plate constantly be compared with the sketch in the note book. If a colony is shaped very irregularly, or if two colonies grow into each other, they are discarded from the sketch. When the colonies have reached the size of a moderate pin's-head they are ready to be transferred into beer-wort.

The pure culture thus obtained is an *absolute pure culture*, because it originated from individual cells the development of which were directly seen. Thus we know that every colony in the places marked o came from one cell. As we had a mixture in our sample, we may expect that the colonies represent the species in the sample, and that they have developed and will develope in accordance with the specific nature of the mother cells.

When the colonies have reached the size mentioned above, the moist chamber is again taken into the culture box and a number of beer-wort flasks are likewise put in. The chamber is now loosened from the slide, samples from each of the small colonies in the gelatine have then to be carried into wort-flasks. For this purpose a number of small infection needles should have been made by cutting pieces of a platinum or aluminum wire, each about one-fourth inch long. One of these small needles is grasped with a forceps and dipped into one of the small colonies, then it is dropped in one of the flasks and left there. In the same way the rest of the colonies are transferred and when what can be used have been disposed of, so that each flask contains a representation from each colony, the flasks are numbered and placed in

*The development of the individual cells was first studied by *E. Mitcherlich*. See *Knapp*, Chemische Technologie II, 1847, p. 273 ; *Schulze*, Chemie fur Landwirthe II, 2, p. 120, 1860, and Report of the (U. S.) Commissioner of Agriculture for the year 1864, p. 517.

the thermostat by 25°C. Afterwards they are examined every day.

We now return to the case in question. We had 32 colonies each of which was transferred into a flask. Two days after this infection there was a vigorous growth in all of the flasks, and, when the microscopic pictures was compared, there were three different forms of cells represented :

1. Round and oval isolated cells in flasks No. 1, 3, 4, 10, 12, 15, 18, 25, 27, 29 (see Fig. 3).

2. Round, oval and pastorian cells in flasks No. 2, 5, 6, 7, 8, 9, 22, 24, 28, 30, 31, 32 (see Fig. 4.).

3. Round and oval cells in short rows in flasks No. 11, 13, 14, 16, 21, 26 (see Fig. 5).

Flasks No. 17, 19, 20, 23 gave a mixed picture, and were, therefore, discarded.

From each of these three series, two of the most typical ones was selected and "freshened up" in new wort for 24 hours three times. After this they were transferred to gypsum and their specific natures determined. No. 1 was *S. cerevisiæ I* (var. easily forming spores), No. 2 *S. Pastorianus III*, and No. 3 *S. cerev. I* (var. not easily forming spores).*

If an infection by moulds or spores of these is suspected, a wort-gelatine plate should be prepared. Here the colonies of cultivated yeast are round or slightly oval, while the colonies of wild yeasts (especially *S. Pastorianus III*), have fringed edges or, when the edges are even, they are somewhat cone-shaped. Colonies of *S. mycoderma* (*M. cerevisiæ*) are deepened in the middle, and thus easily recognized. The moulds (*Mucor*, *Eurotium*, *Penicillium*) appear with their characteristic mycelium and fructification, and cannot be mistaken. The preparation of gelatine plates is described in many bac-

*Since systematic data were given in my paper (Am. Nat. XXVII) they are not repeated on this occasion.

teriological and physiological text-books and laboratory guides.

FRACTIONAL CULTURES.

There is more than one way of making pure cultures; none of these is absolutely exact save Hansen's method described above. We see that Hansen's method is not the same as Koch's plate method, and that the difference is that we are by the former, able to trace the development of individual cells. This, we cannot do by the latter; in bacteriology we see the *colonies* before we see the *cells*.

A pure culture of yeast may be obtained more easily by a *fractional culture* than by the absolute pure culture. The number of cells in one cm^3 of the well stirred sample is ascertained by one of the many counting apparatus existing, and this number we call n. A certain number (s) of cm^3 are then transferred from the sample by means of a pipette, into a flask containing v cm^3 of sterile water. When this emulsion has been stirred, each cm^3 of the water will contain $\frac{n \times v}{s}$ cells. The dilution of the emulsion is carried further until $\frac{n \times s}{v} = \frac{1}{2}$, or there are e. g., 30 cells in 60 cm^3 of water. When this has been done, 25 or 30 flasks with beer-wort are infected each of them with one cm^3 of the diluted emulsion, and thus we have the prospect that as far as can be judged from calculations, half of the entire number of flasks contain each one cell. As the method is, however, not absolutely certain, we run the risk of finding in some flasks no cells, while in others we may find two or more cells. The flasks are placed in the thermostat by 25°C., having been carefully stirred. Afterwards, stirring should be entirely avoided, the cells will then descend to the bottom. The cultures are now looked over daily for about a week. It is evident that the cells have developed in the places where they have been deposited, and thus each cell is represented by a small, white, round spot or flock

at the bottom of the flasks. Such flasks as contain two flocks are discarded, and only those containing one round colony are kept, and further developed. Their contents may be aerated by a slight stirring. The remaining part of the procedure is the same as has been mentioned under the heading of the absolute pure culture. This method may give useful results, but as it is based upon calculations which are to a certain extent uncertain, the absolute pure culture is preferable.

ZYMOTECHNIC WATER ANALYSIS.

There is an essential difference between the hygienic, (chemical and botanical) analysis of water, and the zymotechnic analysis. While the former helps us to find the pathogenic micro-organisms, and their products, as well as such substances as are dangerous to the animal organism, the latter cares only for the organisms that are able to develope in the beer or beer-wort. Many organisms live and develope in water without having the slightest effect upon beer-wort or beer. On the other hand, all organisms that will develope in beer, will also develope in beer-wort. The procedure is : A few cm^3 s. of the contaminated water are disseminated into flasks with wort, and the development is noticed for a term of fourteen days. The preliminary examinations are like those employed by fractional cultures, one germ or cell being introduced in each flask with the water.

ZYMOTECHNIC AIR ANALYSIS.

It is important to know the nature and average number of organisms in the air in and about breweries. The examination of the air is based upon the same principles as that of water, but the organisms are here collected by means of an aspirator. Such an aspirator has been constructed by Hesse (for figure and description, see Salomonsen, l. c. page 505), and serves well for all purposes in this connection. Aeroscopes or aerobioscopes have been

constructed *en masse* since 1859; for these, we refer to bacteriological literature and laboratories. The organisms are collected in water, and disseminated as above mentioned.

In conclusion we shall enumerate the yeast-like fungi that may be found in fermenting fluids; for descriptions, we refer to botanical and bacteriological text-books, such as those of De Bary (l. c.), Fluegge, Zopf (l. c,) Joergensen (Die Mikroorganismen der Garingsindustrie, ed. 3. Berlin, 1893), Ludwig (Lehrbuch der niederen Kryptogamen, Stuttgart, 1892), and bacteriological works by Klein, Woodhead, De Bary, Abbott, Sternberg, etc.

I. *Sacchauomycetes.* Twenty-two species of these are known (see Bay, l. c.); about 30 other organisms have been placed under the heading of this genus; they may retain their place if they are capable of forming spores. For these species, see Saccardo (De Toni) *Sylloge fungorum*, Vol. VIII, p. 919, 1888.*

II. *Torula*, in the sense of Hansen and (exparte) Pasteur. Seven species were described by Hansen (see Joergensen)*, and one was named by Groenlund (*Torula Novae Carlsbergiae*) in 1892.

III. *Saccharomyces apiculatus* was mentioned in the first part of this paper; it is very often found in wine-yeast. It does not belong to this genus, but must retain its name, until its true nature has been revealed.

IV. *Mycoderma cerevisiae* and *vini* were also mentioned above. (Bay, l. c. page 696).

V. Spores of moulds are easily distinguished by their characteristic growth.

VI. *Monilia*, *Chalara* aud *Oidium* are three genera whose species can develope under certain circumstances, into a growth similar to that of the yeasts. But if these fungi which systematically stand near to the moulds and

*S. *Joergensenii* Lasche (See Bay, l. c. page 692) was by Joergensen (the micro-organisms of fermentation, new ed. London, 1893. page 177), referred to *S. exiguus* (Reess) Hansen.

which deserve careful revision in this regard, are studied in pure cultures, it will soon be seen that their nature is different from that of the Saccharomycetes.

It will be seen that Hansen's name is intimately connected with all these questions. During the last fifteen years this eminent investigator has inaugurated a reform in the industrial branches that are based upon fermentation. This principle is to work in praxis with pure cultures exclusively, and through the efforts of Joergensen and other investigators, his methods are now partly or wholly adopted all over the world. His views are at present entering the milk industry, and the manufacturing of wines. Much has yet to be done, but it is seen that the old principle of keeping gardens and fields free from weeds must be recognized also by lower plants, and especially when the successful maintenance of this principle stands behind great factors in the life of individuals and nations.

LITERATURE.

(a). *On pure cultures :*

Hansen: Meddelelser fra Carlsberg Laboratoriet, Vol. II, Part 2; Vol. II, Part 4; Vol. III, Part II. (1883-1892). Transactions of the Laboratory Club, London, 1889. Holm : Medd. Carlsb. Labor. Vol. III, Part 1, 1891.

(b). *On fractional cultures :*

Lister: Quart. Journ. Micr. Science, XVIII, 1878. Klebs : Archiv f. experim. Pathologie, I and Hansens papers.

(c). *On water and air-analysis :*

Hansen : Methode fur Analyse des Brauwassers in Ruck-sicht auf Mikroorganismen. (Zeitschrift fur das gesammte Brauwesen 1888, No. 1). Ueber die zymotechnische Analyse der Mikroorganismen der Luft. (Prager Brauer und Hopfenzeitung, Prag 1888, No. 19).

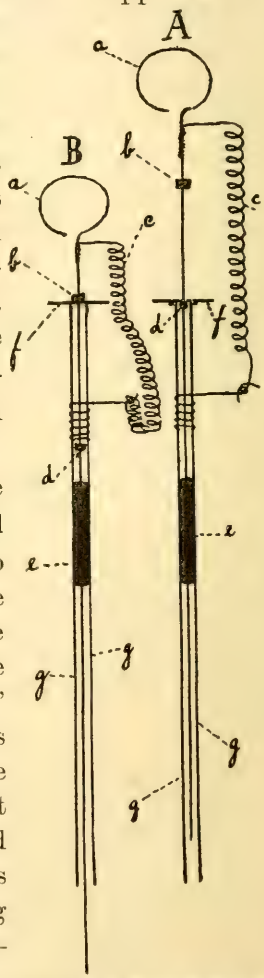
(d). *Historical :*

A. Joergensen : Ueber den Unterschied zwischen Pasteur's und Hansens Standpunkt in der Hefefrage. (Zeitschrift fur das gesammte Brauwesen, 1888). Also : Pasteur : Etudes sur la biere, Paris 1876 (engl. transl. exist-

ing), and historical introductions in Hansen's papers. Belohoubek: Dr. Emil Christian Hansen, eine biographische Skizze. (Zeitschrift fur das gesammte Brauwesen, 1889, No. 24.

A NEW INFECTION NEEDLE.*

The needle itself consists of brass or copper wire. This material is, as a general rule, as good as platinum, provided that it is taken care of, and not allowed to become oxydized. It is passed through the center of a metal disc (f) and its lower part is surrounded by a glass tube (g). The disc can move between two stoppers (b and d); above the former of these, we have the grip (a). This is connected with the glass tube by a spring which under normal circumstances holds the instrument in the position A.



When the needle is to be used, the upper part of the glass tube is charged with cotton. A cotton stopper is also placed in the tube (e). The outside cotton wrapper should have the same size as the opening of the flasks. The general object is to "make a transfer" from one flask to another. For this purpose, the cotton stopper in the original flask is, by means of the left hand, removed from the opening, and in the next moment, the needle is pushed in; the cotton wrapper filling the whole opening and thus prevent-

*The general idea was presented before the A. A. A. S., Sec. G., at Madison, in August, 1893.

ing infection from the air. The needle can now be moved in all directions in the flask, or test tube. When the grip is pressed downwards, the needle passes the opening of the glass-tube, and it can then be "infected" or touch a colony, sporangium, etc.; after this the apparatus is taken out, and replaced by the original stopper which has been in the meantime, flame sterilized.

While the stopper of the flask is removed the cotton wrapper of the needle should pass the flame. The latter is then inserted in the new flask and the inoculation made. After this the whole needle may remain in situ, or it is taken out in the same way as it was taken out of the first flask. The glass tube alone may remain in this case, the needle itself is taken out and the stopper (e) is left as a filter.

The transfers may be made in the Hansen box. The needle can be used for many purposes, e. g.: secondary introduction of medium (when the glass tube is left after inoculation), its manipulation is easy, and the sterilization carried out without difficulty. A better form may be constructed on the basis of the same idea. At any rate this is the first attempt of abandoning such rude instruments as a glass rod or an unprotected piece of wire as instruments for handling micro-organisms in the vegetable kingdom.

Bacteriological Laboratory, Iowa State Board of Health, Dec. 24, 1893.

Aeration of Tissues and Organs in Mikania and Other Phanerogams.

By W. W. ROWLEE,

ITHACA, N. Y.

The author's investigations were undertaken in order to ascertain if possible the function of certain ærotropic roots and consisted of an examination of the details of structure of these roots and especially those produced by *Mikania scandens*. This plant belongs to the order

Compositæ and is closely related to the Thoroughworts. Attention was first attracted by the peculiar appearance presented by these roots in summer and autumn. They grow in mud and water and, at the above mentioned seasons, produce an extraordinarily extensive root-system. Many of the aquatic roots have an unmistakable tendency to grow up to the surface of the water; the roots, growing in mud or even transplanted into a dry place, still show a tendency to grow upward.

The plants studied grew along the shores of the Oswego river in northern New York. It is associated there with *Decodon verticillatus*, the water-loving Polygonums, and other shore plants over which it grows in great profuseness. Its clusters of flesh-colored flowers are produced in August and September.

A careful study of the structure of the root and also of the literature bearing upon the subject leads to the conclnsion that the function of these roots is to aerate the submerged parts of the plant.

If respiration be one of the primary functions of organisms and no one will assert that it is not, it is to be expected that modifications to increase the facility and certainty of its accomplishment will be found among the more highly organized plants. The modification of organs to secure the best conditions for aeration is the direct result of an evolution of forms best adapted to live. Indeed one might justly say that respiration is the first of the functions. The first signs of germination in a seed is the enlargement of air-containing spaces between the cells. In some cases these spaces become comparitively large before active cell division begins. During the succeeding life of the plant, where the massiveness of tissue or the lack of aeration in the surrounding medium makes it advantageous, provision is made for aeration either by intercellular spaces without any regular organization of tissues, or by a special tissue organized to in-

crease the ease with which air is supplied and regulated.

The part of the plant which is to be aerated is that which is alive and active. Much of the tissue making up a plant is not alive. It is the active protoplasm of living cells that is the seat of life in the plant and it is that substance which must be both aerated and supplied with water. The protoplasm of a matured active cell usually assumes a definite form and arrangement of parts within the cell-wall. The protoplasm constantly changes its form as well as its chemical and physical properties but nevertheless certain parts may be distinguished. These are the nucleus and more or less clearly differentiated ectoplasm. The latter lies in very close contact with the cell-wall in the living cell but contracts from it under the influence of certain reagents. Sometimes the ectoplasm of adjacent cells are visibly connected. This continuity of protoplasm occurs in the cortical tissues of many woody plants and in the cribrose cells of the fibro-vascular bundle. It has been asserted that the protoplasm of all adjacent cells is continuous, the ectoplasm of adjoining cells being connected by invisible threads. Some have gone so far as to say that this can be demonstrated. The irritability of plants, and the power which plants have of absorbing and reforming their walls, can perhaps best be explained upon this ground.

It is to the ectoplasm and to the nucleus that whatever the cell needs to live upon, must come. It was once believed that cutinized cell walls were impervious to gases. This view is now abandoned. Graham, an English physicist and chemist, first formulated the laws upon which the modern idea of the dialyses of gases permeating a rubber membrane are based. Barthelemy hastened to apply these laws to plant membranes such as the epidermis of the leaf or stem. He even intimated that stomates and other openings in cutinized membranes were of minor importance to the plant in its exchange of gasses.

Sachs however in his extended investigations, came to the conclusion that the stomates are the principal agents in the exchange of gases. The tendency of recent investigations is toward the establishment of the idea that exchange of gases takes place between the internal air chambers in the plant and the surrounding air both by effusion through the stomates, lenticels etc., and also by diffusion of gases through the cell-walls even when they are cutinized.

The air-spaces in the body of the plant then are entitled to much more attention than has beeu accorded them by investigators.

In submerged plants and plant parts, the abundance and large size of intercellular spaces has long been known. They are abundant and of large size in the roots of the plant in question. They are especially abundant in the aerotropic roots which are produced in autumn in such abundance. They form longitudinal channels through them and into these channels the external air finds access by diffusion.

A condensed statement of the conclusions reached, (the details of the work may be found in the Proceedings of the Am. Micro. Soc. for 1893) is as follows:

The internal structure of the roots is characteristic of the structure of submerged organs of aquatic plants, as set forth by M. Constantin. (Ann. sc. nat. XIX Bot. p. 287, 1884.) The reduction of the xylem and the increase in capacity of intercellular spaces, are conspicuous features in the submerged organs.

When grown in the garden for a period of years the organs tend to develope a more perfect xylem and at the same time reduce the size of the intercellular spaces.

Gases may pass from one compartment to another by effusion as well as diffusion. The connecting openings between spaces are not as conspicuous in the root of *Mikania* as they are in the organs of some other species,

for example *Menyanthes*. Gases to pass into the organ must traverse a living continuous membrane. This they do by diffusion.

Intercellular spaces occur in the phleom of the radial bundle. These spaces have large cells lining them, the nuclei of which have an affinity for the side of the cell which borders the air space. This is presumably because the conditions of life are better there.

Cornell University.

Centering Device for Turn-table.

By E. E. MASTERMAN.

NEW LONDON, OHIO.

All turn-tables should be self-centering or have a device for centering which is not mechanical. Centering has become an absolute necessity for neatness where round cells are desired. It is not an easy task if you have no means for centering except the eye. I will give a method for marking the top of a turn table which I have used for some years. Any one can use it, and those who have no centering device are urged to try it. With a little patience and neatness, centering can be nicely done and it will save much valuable time, while it costs nothing. The device will be readily understood by refering to the figure and observing the directions.

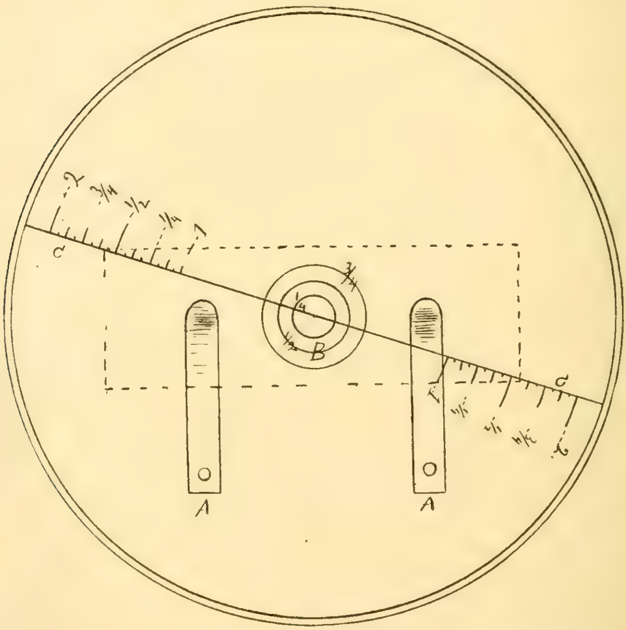
DIRECTIONS FOR MARKING THE TABLE.

1. Draw a diameter in relative position to the clips AA, as in the drawing, p. 50. Be very accurate, so as to have it pass through the center.

2. Draw several concentric circles near the center as shown in the figure, p. 50. Three is usually enough—one-fourth, one-half and three-fourth inches. These are to be used as guides for the circle or rings which you make upon the slide. The fractional numbers denot the size of covers. (See B in the figure.)

3. Draw a scale each side of the center, at CC; begin one inch from the center, or nearer if you wish. Be sure that both sides are exactly alike. Make the notches 1-16 inch apart. Use a very sharp-pointed pair of compasses to mark with, but if you have none, paste a white paper over the top of your table and mark with a pen and ink. Some prefer the paper.

To use the device, place a slide upon the table as indi-



cated by the dotted lines, with two of the corners upon the scale, being always sure that the opposite corners are equally distant from the centre. For example, if the slide measures three inches from one corner to the opposite corner, place the opposite corner at 1½ on the scale. Press down the clips firmly, and you are ready to proceed.

Be accurate; take a little extra time at first until you become accustomed to use it.

Radiolarian Shale from Manitoba.

BY FRED'K B. CARTER,

MONTCLAIR, N. J.

In the American Monthly Microscopical Journal for April, 1893, there was a description of some Radiolaria from a locality in Manitoba, and what made the article most interesting to the student of the Radiolaria was the fact that of the 16 species there mentioned 13 were new, and the other 3 were not found in the Barbadoes earth, at least they have not been found as yet. And as specimens of this shale, according to the writer of the article were found to contain large numbers of well-preserved Radiolaria the presumption is that many more species in the Manitoba material will be found to differ from those in Barbadoes. The material therefore is worthy of the closest study, especially as it comes from our own continent. A few words as to the best method of cleaning it may not be amiss now that some of the material is at hand.

At first sight, it seems very unpromising, especially to one who has become accustomed to deal with the Barbadoes earth. For in the first place it is very dark in color as compared with the latter and in addition it is much harder, breaking like a piece of slate. You can rub off some of the material of the Barbadoes earth between your fingers, in fact you can hardly handle it without finding the fingers covered with a white dust; but this shale is smooth and no amount of rubbing will free any of it. But in reality it is by no means as bad as it looks and can be reduced without much difficulty by means of the ordinary carbonate of soda. The amount of soda used, however, should be greater than in the case of the Barbadoes earth, two or three times as much soda in size as the material to be reduced. Take a porcelain or stone lined saucepan holding about a quart and let it be

two-thirds full of water. Then for a small cube of the material put in two or three equal sized lumps of soda and let the water boil briskly for half an hour or more. Then wash thoroughly in four or five changes of water, each time filling up nearly to the top and rotating the saucepan by the handle and allowing to settle for from one to two minutes. Now pour off the water until the saucepan is only about one-third full, add a teaspoonful of pure nitric acid and boil for two or three minutes. Wash again thoroughly to remove the acid, letting the material settle a little longer each time than at the first washing. Pour off well down and fill the saucepan two-thirds full again; add a lump of soda, and boil for five minutes. Wash again thoroughly to remove the soda. Pour off water well down and transfer material to a tall thin glass or ordinary tumbler and let it settle thoroughly. Then pour off and transfer material to a porcelain evaporating dish. Remove as much water as possible and add a good quantity of pure nitric acid. Put in a few large grains of sand to prevent bumping of acid, cover with a saucer, right side up, and boil over an alcohol lamp in a retort stand for three or four minutes. Wash again several times in tall glass or tumbler, allowing it to settle well, and then boil again in the evaporating dish with a small piece of soda. Then wash to remove the soda and the material will be found quite clean so clean at least that when mounted in Canada balsam, as it should be, the forms will be transparent enough for study. But if the result is not as satisfactory as one could wish all that is necessary is to boil again in pure nitric acid and then in soda. And if there is an excessive amount of foreign material mixed with the Radiolaria most of it can be removed by pouring the cleaned material into a test-tube and allowing it to settle not longer than thirty seconds. After pouring off, the sediment will be found to be almost entirely Radiolaria.

The piece that was sent me for examination was the merest bit, so small that if it had been Barbadoes earth one would not have taken the trouble to clean it, and yet it yielded a good many forms and a number that were strange to me, which may serve to show how rich the material is. Of course, under the circumstances, I was not disposed to waste any of it by excessive washing and there was considerable foreign material mixed with the final settling and the forms were not as clean as they would otherwise have been, but I am convinced that the process given above will yield good results, though it may need to be supplemented in order to render some of the bulkier forms perfectly transparent. Sometimes, indeed, it seems almost impossible to get all the forms free from the foreign material lodged in the skeletons. For example, there is some material which was sent me from Barbadoes by Prof. G. Frith Franks, a rolled pebble from the shore near Jois River, which I have boiled and boiled in the strongest acids without being able to remove all of the dark red foreign material from the shells. But those forms are sufficiently clean for study.

I may add that it will be well to break up the Manitoba material into small and thin pieces before subjecting to treatment.

On the Development of the Continental Form of Microscope Stand.

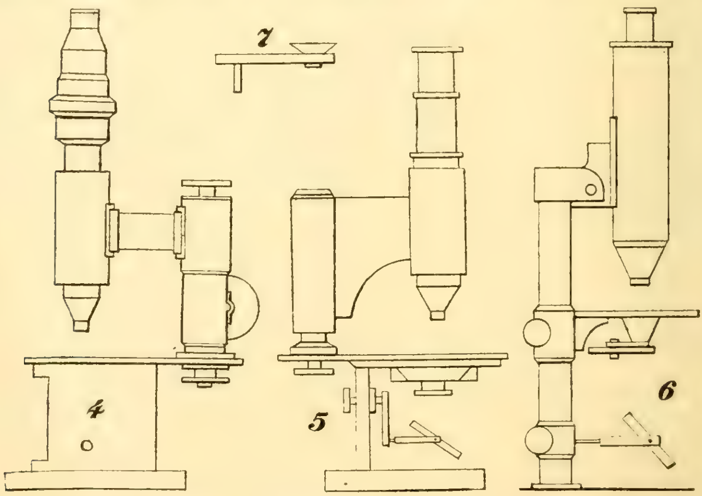
By J. B. NIAS, M. D.

LONDON.

Concluded from January, page 27.

In Fig. 4 is represented the stand as patented by Trecourt and Oberhaeuser, after a cut in Dujardin's article and the drawings of the patent; specimens of the manufacture, however, which I have seen correspond more closely to the design of Fig. 2. Certain features which

suggest themselves more particularly to the anatomist as indispensible, have been suppressed, such as the outer tube for swinging the body to one side, and the numerous sockets for stage forceps; and the rotating diaphragm has been replaced by a tube moving vertically through the opening in the stage, by the aid of a lever at the side, so as to take from above the achromatic condenser of Dujardin patented at the same time, or a set of cylindrical diaphragms, a feature of which Strauss-Durckheim expressly objected as necessitating the disturbance of



the preparation on the stage. A rack-and-pinion coarse-adjustment has been added, and the milled head of the fine-adjustment removed to the top of the pillar.

This instrument came into great favor, much aided by its cheapness, and the goodness of the objectives; and and was exhibited at the sitting of the Academie des Sciences on February 13, 1837.

On the occasion of Nachet exhibiting at the Academie des Sciences an apparatus for oblique illumination by means of a prism on June 14, 1847, ten years later, Oberhaeuser, now in the business by himself, announced

to that body that he had modified his stand so as to admit of the use of the mirror for that purpose, the idea having been suggested to him from England by a Mr. Abraham, of Liverpool, and a patent was taken out for this improvement also, though not till two years later. Fig. 5 represents this second stand, which has now arrived at its modern form. The sides of the drum have been cut away so as to leave a flat pillar, and the round foot has been modified, to correspond, into a horseshoe form. The mirror, borne by a swinging arm, is adjustable vertically by means of a clamping screw. The stage rotates, as before, with the body, but is now made square, and the diaphragms and condenser are carried by a sliding substage. A hinge for inclining is the only subsequent addition to this model; and the smaller stands with which we are more familiar in this country in the hands of students, are derived from, and posterior to the invention of this larger one, which continues to be made by every Continental optician at the price of 10*l*. or 15*l*.

In Fig. 6 and 7, I have added, for contrast, the type of microscope current at the time of the introduction of the Oberhaeuser stand; it is that made by Chevalier for Dujardin, and with which he worked. Like others of the period, it is fixed for use by screwing into the lid of its cabinet, and the focusing is effected by movement of the stage. In the present specimen the compound body can be lifted off the pillar, to be replaced by the arm carrying a simple lens shown in Fig. 7, and by means of a hinge and the interposition of a right-angled prism above the objective, it can be used in the horizontal position. It is, in fact, a simplified form of the "Microscope Universel" of Chevalier. Stands of similar form were made by Plossl and others at the time, and were equally superseded by the new pattern.—*Journal of the Royal Microscopical Society.*

Collecting and Studying Parasitic Insects.

By HERBERT OSBORN,

AMES, IOWA.

While the common insect parasites affecting birds and mammals are usually looked upon as rather disgusting and uninteresting creatures, they may become to the microscopist objects of no little interest. They possess numerous points of structure adapting them to a very peculiar mode of life, some of which are remarkably interesting but since they can only be observed with considerable magnification, they are of course unseen by any but a microscopist. The collector need only to conquer a dislike caused by the habits of these animals to find in them sources of interesting study. They may be collected from both birds and mammals, the groups which are referred to here occurring only on warm-blooded animals. For preservation, they may be put in vials of alcohol or mounted in Canada balsam on slides and where specimens are abundant, it is advisable to preserve some by both of these methods. Fresh specimens may be mounted in balsam and can be studied at once, but the balsam will cloud later and remain so for perhaps some weeks, so that if it is expected to study the specimens for some days after mounting, it is better to first dehydrate them with alcohol and clear with oil of cloves before using balsam.

Alcoholic specimens may at any time be mounted in balsam, clearing with oil of cloves, chloroform or other clearing agent. Frequently specimens can be collected from the skins of dried birds or mammals in museums, and will frequently be found in excellent condition at least for the study of the more important parts in identification. Such specimens will often exhibit air spaces which interfere with their study and injure the beauty of the mounts, but I have found it quite an easy matter to remove these air spaces by putting the specimens under

cover glass on a slide and filling with benzole which is alternately heated and cooled adding fresh benzole to supply the loss from evaporation until the spaces are seen to be entirely filled. Then balsam in benzole is added at the edge of the cover and the mount left to finish. The specimens in vials or on slides should be labeled with at least the name of the host animal and will naturally receive the date and name of the collector in addition. The record of dates for specimens in this group have however little importance as regards the life history and development of the insect.

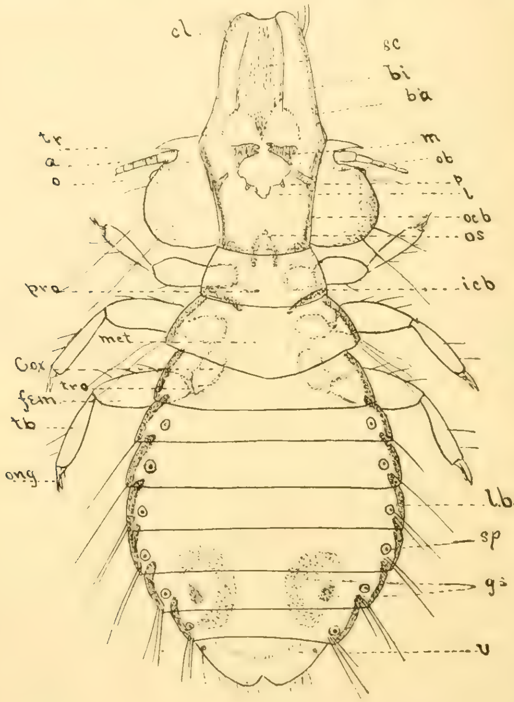
These parasites are contained in two groups, the Pediculidæ and the Mallophagidæ.

The first group forms a division of the Hemiptera and has frequently been made a separate suborder (*Parasita*) and placed as the lowest of the Hemiptera. This however is based only on the simplicity of structure due to degradation and is clearly unwarranted on any basis of philogeny.

The second group was formerly placed as an aberrant division of the Hemiptera, but is now recognized as having its affinities with the lower Mandibulate insects and related to the *Psccidæ.* The Pediculidæ are found only upon mammals while the Mallophagidæ appear upon mammals and also upon birds. The Pediculids are quite constant as regards the species of animal which they infest ; so much so that ordinarily species may be determined by simply knowing the animal from which specimens have been taken, nevertheless the student will desire to know something of the characters which distinguish the different genera and species. They are mostly soft bodied, have simple tubular mouth parts, the antennæ are simple, usually of five joints and the legs are frequently modified in different ways and also adapted for clasping, the claws being strong and opposed to prominences upon the tibiæ. In some cases there is a special structure for

clasping, noted especially in the hog louse in which the end of the tibia is a protractile disk which presses against the hair. The abdomens are large, the spiracles prominent and in some species there is a peculiar brush-like organ on the under side.

The function of this organ is unknown and hitherto no suggestion has been made as to its office. It would seem



probable that it serves to retain the abdomen in a proper position for the deposition of eggs, the hair to which the insect is attached being held between these two brushes.

The characters most used in classification are, the form of head, the number of joints in the antennæ, the form of sternal plate and of abdomen, the structure of the tarsi and the special appendages on antennæ, legs and abdomen.

The Mallophagidæ are classified mainly on modifica-

tions of head, antennæ, palpi and tarsi, the more important parts being illustrated in the accompanying figure of *Docophorus* adapted from Piaget, (*Les Pediculines.* Supplement Plate 17.) The abbreviations are as follows: Cl. clypeus, sc. clypeal signature, bi. internal band, ba. antennal band, tr. trabecula, a. antenna, o. eye, m. mandible, ob. ocular band, pl. labial palpus, l. labium, oc. b. occipital band, os. occipital signature, icb. intercoxal band, pro. prothorax, met. metathorax, cox. coxa, tro. trochanter, fem. femur, tb. tibia, ong. onglets, lb. lateral band, sp. spiracle, gs. genital spots, v. vulva.

In another paper may be given a key to the species found on our common animals for the convenience of those wishing to determine their finds. The writer will be pleased also to determine specimens sent him for the purpose.

A Homogeneous Objective Condenser.

BY WILLIAM LIGHTON,

OMAHA, NEBR.

The attention of the microscopist is now more than ever engrossed with the important part borne by the condenser in contributing to satisfactory results of his work. A good condenser is now no more a mere adjunct: it is a necessity. For many years I have been experimenting in this direction, with results which I wish here to partially outline.

In *The Microscope* for October 1891, I described some of the effects obtained by the use of a homogeneous immersion objective as a sub-stage condenser. That was but the beginning. I have since carried forward experiments looking toward the perfection of a homogeneous immersion condenser, and with the valuable aid of one of England's best opticians have attained the desired result. I have now before me the first instrument constructed on

the new formula, and find it to be in all particulars in conformity with the end sought to be attained, a condenser capable of as delicate and accurate focusing as a high-grade objective under any possible use of light, perfectly corrected for both spherical and chromatic aberation, and having a numerical aperture of 1. 30. Workers with the Abbe form of condenser realize the necessity for and inconvenience of constantly changing the focus of the apparatus, as the method of lighting is changed from central to oblique; with the new condenser this is obviated, the focus being constant. An Iris diaphragm is used with the new condenser, and by the use of an adapter it can be accommodated to any form of sub-stage mounting in use. The name of homogeneous-objective-condenser has been given it, because of the fact that, owing to the perfect correction of the optical combination, it can be used in the body-tube of the microscope as successfully as any other homogeneous immersion objective. It thus serves a double purpose. The instrument in my possession has been corrected for thin and medium glass slides, and when used as an objective it is better to invert the slide upon the stage.

In experimenting as to the relative efficiency of the new form and the Abbe condenser, I have selected as a good test a mount of *Amphipleura pellucida*, in Smith's medium, using a 1-12 Lietz apochromatic homogeneous immersion objective. With the use of the Abbe, I only secured a resolution of the transverse lines, while with the new form, with the same handling as was given the Abbe, the longitudinal markings were well shown. When using small stops for the purpose of bringing the light to the extreme edge of the objective, the contrast between the work of the two condensers was most strongly marked.

Various makers have heretofore furnished achromatic condensers of 1. 0 numerical aperture, but this is too small to admit of the best results being obtained with

fine objectives, and these condensers are open to the additional objection mentioned in the fore part of this article, they are not susceptible of a sharp focus. Powell & Lealand have furnished a fine condenser of 1.40 numerical aperture, but at a high cost, while the new form can be supplied to workers at a low price. All will be thoroughly tested.

I shall be glad to correspond with any who wishes further information upon the matter.

2511 SEWARD STREET, OMAHA, NEBR.

LETTERS TO THE EDITOR.

NOTE.—*This column is open to all correspondents who write upon the topics enumerated under "Problems," or who give other information of interest. The fact that a problem has been answered once need not deter our friends from making additional comments. To facilitate reference, correspondents should cite the number as well as the page on which have appeared letters and queries to which reference is made. The editor is not responsible for the views of others published in this periodical.*

Rush Medical College.—Replying to your note, I would say that all of the students have systematic training in the use of the microscope, in the branches of Histology, Pathology and Bacteriology, for which we have new and thoroughly appointed laboratories. Each man is supplied with a complete outfit, including a high grade microscope. Each of these courses extends through eight weeks, and includes between thirty-two and forty-eight hours practical work in the laboratory which is as much time as seems judicious for undergraduates to spend upon these subjects. E. FLETCHER INGALS, *Registrar*.

CHICAGO, *January* 18, 1894.

Tariff on Books.—About the tariff on scientific books I am wholly and cordially opposed to it. It seems to me, under the existing circumstances, of very small and doubtful advantage to both authors and publishers, while the importance of improved education is universal and urgent and is recognized as a public burden by the heavy taxation endured without objection for its relief. Even assuming the wisdom of protection as a general policy, it seems to me more important at present to encourage the education of the many who are deficient in it, than to patronize with artificial stimulus the few who can and will

write and publish, any way, more books and journals than the rest have opportunity to study to advantage. R. H. WARD, Troy, N. Y.

Spores in Syphilitic Blood.—Dr. E. Cutter, writes: "I have for the first time in a medical college demonstrated at the Boston College P. and S. on the screen the saltating spores of syphilitic Blood."

EDITORIAL.

The Microscope in School.—At Mr. Moody's school in Northfield, a pupil says that they examine the blood of his pupils with the microscope as a part of the instruction, which is a move in the right direction. The time is coming when microscopes will be as common as pianos and organs in houses. If pupils of preparatory schools learn how to study the form elements in blood, it is high time that the medical profession see to it that they are not left behind.

A Chance to Use the Microscope to Save Cattle.—Yesterday January 18, seventeen fine cattle were killed on Ex-Vice President Morton's farm. We have not learned the results, but if the morphology of the blood in tubuculosis is the same as in man, it is possible to tell from the blood whether the cows are tubuculous or not. Some recent trials of this kind came out wonderfully correct as verified by the post mortem examinations.

MICROSCOPICAL APPARATUS.

A Big Microtone.—At Chicago was shown by the University of Pennsylvania a giant microtome used for cutting microscope sections through the entire brain. The object to be sectioned is fixed at the end of a very heavy lever and allowed to sink down upon the edge of a broad knife, the blade of which is parallel to the side of the lever. The section as it comes off is caught on a sloping sheet of paper.

Wax Models of Microscopic Objects.—A method of making models in wax has been devised as follows: A camera drawing is made of each section in the series. The paper, which is very thin, is then attached to a sheet of wax. For an enlargement of twenty-five, the wax must be twenty-five times as thick

as the section just drawn. The edges are trimmed to fit the paper outline and the successive layers of wax are put together in the same order that the sections occupied before being cut.

MICROSCOPICAL MANIPULATION.

Mayer's Carm-alum.—This is a new carmine stain invented by Dr. Paul Mayer. He uses carminic acid instead of commercial carmine. In Leipzig carminic acid costs only eight cents per gramme.

Carm-alum is a dark-red liquid with intense staining power. It gives a beautiful stain to objects that have been fixed in pure osmic acid. Alum carmine will not do this. It tinges the protoplasm slightly as well as the nucleus. Its use in the case of objects mounted entire is not recommended because it may overstain and render them opaque.

Mix together in a suitable vessel one gramme of carmine acid, ten grammes of ammonia alum, 200cc. distilled water. Heat to a boiling point, dissolving the ingredients, after cooling pour off or filter the liquid. It is now ready for use. A small crystal of thymol or of some other antiseptic may be added to keep it free from organisms.

The application of this stain to protozoa, etc., has been described in *Natural Science* for August, 1893, p. 114.

Hermann's Fluid.—This differs from Flemming's fluid by replacing the 1 per cent chromic acid with 1 per cent platinum chloride. It is made as follows:

Platinum chloride, 1 per cent	15 parts.
Osmic acid, 2 per cent	4 parts.
Glacial acetic	1 part.

For example, take one of the sealed glass tubes containing a gramme of osmic, as it is commonly sold, and break it in a bottle of 300cc. capacity; pour on it 50cc. of pure distilled water, 12½cc. glacial acetic, and 187½cc. of 1 per cent solution of platinic chloride. Shake and put aside over night. When the osmic has dissolved the mixture is ready for use.

DIATOMS.

The Grandest Collection on Earth.—The British Museum has purchased the great collection of 30,000 slides of diatoms

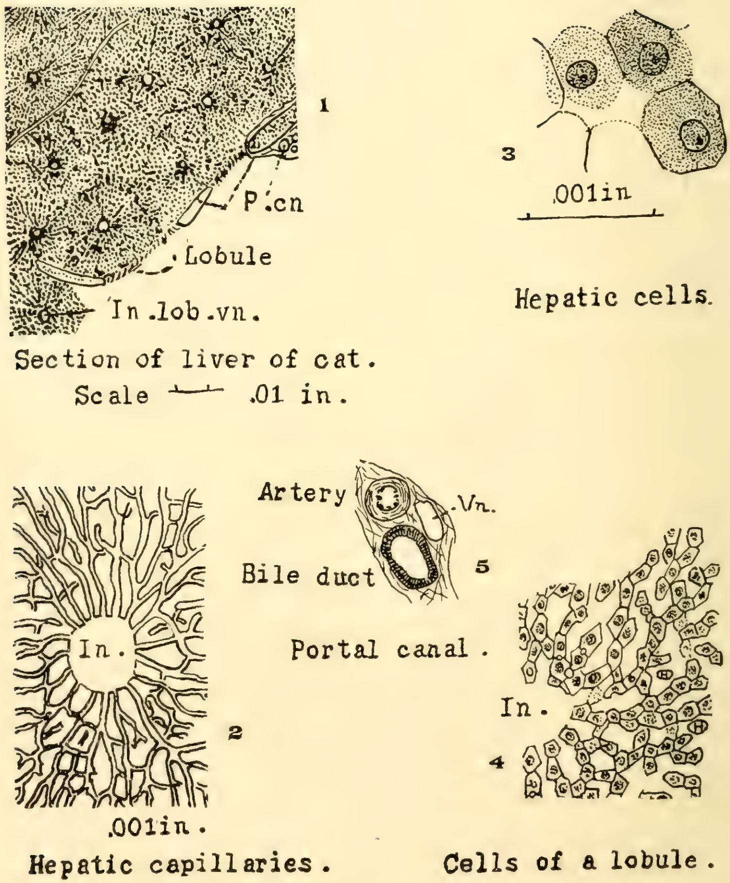
made by Mr. Julien Deby. It is indexed, and accessible to students in the botanical department. The Museum previously possessed Greville's, William Smith's, Gregory's, O'Meara's, Ralf's and other collections.

MICROSCOPICAL SOCIETIES.

The Sphinx Society, Atchison, Kans.

Jan. 11, 1894.—A. H. Lamphear, M. D., presented a paper on "The Germ Theory of Disease." After dwelling briefly on the history of the subject Dr. Lamphear presented the three theories that have had the most following, *i. e.* the chemical theory, the germ theory and the bioplast theory. The various forms of infectious diseases such as anthrax, tuberculosis, diphtheria, yellow fever, etc. in which and for which a specific germ has been isolated were more particularly considered. Examples of tubercle bacillus, anthrax and gonococcus, were shown under the microscope. Also photographs of the same were exhibited; also agar cultures of microbes from the mouth, water and etc. The discussion that followed the reading of the paper was especially interesting and instructive. In the discussion various theories on immunity were presented. One member offered it as his view that in looking for the cause of disease we should not loose sight of the chemical influences while championing the germ theory. We would find a truer explanation of the disorders that arise by remembering that all cells, whether bacterial or of the animal structure, give rise to poisonous by-products in their disintegration, and that it is because of the absorption of these ptomains, toxalbumins, etc., and their deleterious influenc, especially on the nervous system, that diseases arise. Thus disorders may arise within the system without the entrance of bacteria of any kind, and these disorders may resemble very closely the disturbances engendered by actual bacterial growth, because the nitrogenous poisons produced may be almost identical in the two cases. Viewed in this way the chemical theory is deserving of closer attention. The officers of the club for the following year are as follows:

Pres. E. B. Knerr, Vice Pres. C. S. Hull, Secretary, J. H. Glotfelter.—*Reported by E. B. Knerr.*



THE LIVER OF A CAT.

THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. MARCH, 1894. No. 3.

Studies of the Histology of Various Mammalian Tissues.

BY HENRY L. OSBORN,

ST. PAUL, MINN.

WITH FRONTISPIECE.

PREFATORY NOTE.—It has seemed desirable to print in the pages of this Journal a series of articles with illustrations describing minutely the cellular structure of some of the tissues of the highest vertebrates. The intention is to prepare a series which by the help of the text and the cuts will make it possible for anyone who has a good microscope with ordinary range of magnifying power to distinguish in sections prepared with only ordinary methods and care, the essential cellular structure of the more familiar organs. My object in this is to furnish persons, intending to study human physiology, with figures drawn directly from average sections such as they can make with reasonable care, to the end that they may by studying these cuts and sections better acquire the practise needed to teach one to interpret directly from nature instead of being confined as so many are to the plan of resorting to a manual of histology if they desire to learn the cellular structure of organs.

TECHNICAL METHODS OF PREPARATION.—All of the sections herein figured or most of them have been prepared by hardening in alcohol. The finest histological work requires that more careful hardening be practised than is possible with alcohol, but beginners in histology, or any others who are not in search of minuter cytological and

nuclear details, will find that alcohol properly used will give, in a majority of tissues and especially in glandular tissues, entire satisfaction. The tissue should be placed, as soon as possible after removal from a recently killed animal, in ten times its bulk of thirty per cent alcohol, after an hour it should be changed to the same ratio of bulk of fifty per cent alcohol, after another hour it should then be transferred to seventy per cent alcohol. It should be cut into small pieces before the alcohol treatment, into blocks of half an inch on a side, and should be changed from the seventy per cent alcohol after twenty-four hours to another bath of the same strength. It will then keep indefinitely or can be imbedded and cut at once. Staining in bulk and imbedding by either the celloidine or the paraffine method and cutting and mounting are performed in the usual way.

THE LIVER OF A CAT.—The liver furnishes a very good example with which to begin in the study of the cellular structure of an organ. As a whole the organ lies in the anterior, or in man the upper, part of the abdominal cavity. It is a large dark structure just above the stomach and just beneath the diaphragm. In a recently killed animal, as it is cut, a large amount of very dark "venous" blood flows from the cut surface. Its anatomy is too familiar to require more than the merest summary. A great duct runs from the organ into the small intestine, this is the "common bile-duct, it comes directly from the "gall-bladder" but indirectly from a multitude of lesser ducts which lie in the areas between the "lobules" of which the mass of the organ is composed, [see Fig. 1, P. cn] in a passage called the "portal canal." This same passage between the lobules is the location also of arteries and veins, [cf. Fig. 5]. Besides the "bile-duct" which is an outlet passage for the secretion of the organ, there are also two sets of blood-vessels which enter the organ on its posterior side, these are the "Hepatic Ar-

tery," from the aorta and the "portal vein," from the capillary system of the intestinal tract and the hepatic vein leaving on the anterior side. The vessels as they enter the organ begin to divide into lesser and lesser vessels, and this goes on till they are ultimately minute "arterioles," such as is seen in Fig. 5, in the portal canal, also this is one of the vessels seen at P. cn. in Fig. 1. These vessels of the aortic and portal sources at the time they are ready to pass into capillaries are found in the areas between the "lobules," which compose the substance of the liver. In Fig. 1, a view of the appearance of a section only slightly magnified is presented. As the accompanying scale shows, the enlargement is here only about eighteen diameters. In such a view as this a section will appear to be made up, indistinctly, of hexagonal portions each with a hole in the centre and here and there between the hexagons a vascular-looking passage will be seen. Subsequent study with the higher power will prove that these areas are the cellular tissue of the organ and the fact that sections in any plane will give the same appearance, prove that the organ is composed of polygonal blocks, each one a system of cells with an arrangement like that of all the other blocks. The central hole is called the "intra-lobular vein." By applying the scale to the measurement of a single lobule it is found to be about three one hundredths of an inch in diameter. We thus learn that the coarse anatomy of the cat's liver is a great number of masses called lobules, each one about a thirtieth of an inch in greatest diameter, that these are supplied with blood from the aorta and from the portal vein, that the organ is drained by the hepatic vein which flows off into the right side of the heart, that the secretion of the organ is drained out into the small intestine.

CELLULAR STRUCTURE OF THE LIVER.—To follow out the distribution of the blood vessels an injected specimen is needed. Injected specimens that are sold by the deal-

ers generally are good for the mere purpose of showing the capillary vessels but they do not show the cellular structure at the same time as a rule. An injection should be made with a gelatine mass which will harden in alcohol just as the cells do, and from such a section the relation of the cells and capillaries can be readily made out. Figures 2 and 4 were both of them drawn from the same section, one of them being drawn from the capillaries the other from the cells. The former is a view of the converging capillaries derived from the hepatic artery and the portal vein which as already shown are located on the periphery of the lobule, the capillary vessels converge toward the center of the lobule where all meet in a vein the "intra lobular vein" [In.] which is the beginning of the hepatic vein. In Fig. 4 the cells for a short distance from the center are drawn to show their relation in the lobule. This shows how much intercellular space there is; it is in this intercellular space that the capillaries are located, and as the blood moves along in these capillaries the cells have an opportunity to take from and give to the moving stream. The spaces between the cells also lodge the ultimate terminal twigs of the bile ducts, but these cannot be seen unless the liver has been especially injected through the bile duct.

THE HEPATIC CELLS.—In Fig. 3 I have shown in detail three of the hepatic cells. The cells are destitute of a distinct wall and their outline is bulging and rounded where it is not in contact with a neighboring cell. Each cell is a little less than a thousandth of an inch in diameter, it has a conspicuous circular nucleus, darker in stain and this has a definite circular membrane, and contains a single minute spherical nucleolus. The protoplasm of the cell stains readily and deeply. The intercellular spaces in thin sections are clearly seen they are not readily distinguishable if the sections are thicker than the cells.

POINTS NOT SHOWN.—Besides the points referred to already all of which are shown except the bile capillaries, the liver is supplied with nerve fibres. These are not distinguishable from the connective tissue surrounding them, but it is known that both medullated and non-medullated fibres are given off to the liver from the coeliac plexus as well as from the pneumogastric nerve and it is probable that these are distributed both to the arterioles "vaso-motors" and to the glandular cells [see Landois and Stirling, 324].

CONCLUSION.—This is not the place for any extended discussion of the use of facts thus learned of the liver in aiding toward an understanding of the physiology and pathology of the organ, but I may perhaps allude to the matter. The hepatic cells as well as the other living cells in the organ are subject to the same laws of life as organisms in any situation, hence conditions of food, temperature, alternation of rest and work, and all the conditions which affect life must affect these living cells. The cell is so situated that it is entirely at the mercy of its surroundings and unless these are hygienic the cells must suffer. If the liver is in a healthy state, and sections are made of it after a meal especially one in which an abundance of starchy food has been taken the cells are seen to contain granules of glycogen, before such a meal the cells are as shown in our figure composed of protoplasm wholly. The secretion of other than the normal material as for instance of fat or of pigment if it was to take place on an extensive scale would cut out a large number of cells from the aggregate of workers in the organ, if cells should die and their places be taken by foreign cells this would have the same effect, the vascular system might become defective so that the cells would not receive the needed food or oxygen and thus a series of difficulties are possible on this line, while if the temperature of the organ rises too high here again are

difficulties against which perhaps the cells are unable to contend. Disease of the organ then may arise from any of these causes and one if inaugurated may bring on the other, in any case it is because the normal conditions of cell life are interfered with when the organ is an "unhealthy state."*

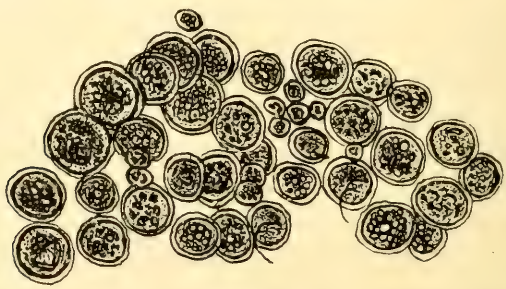
BIOLOGICAL LABORATORY of Hamline University, Jan. 27th, 1894.

Red Snow as Seen by Means of the Microscope.

BY ARTHUR M. EDWARDS, M. D.

NEWARK, N. J.

Having received from Prof. Harry Fielding Reid a specimen of *Hœmatococcus* from Tidal Inlet, Glacier bay, Alaska, which he desires me to report upon, I have to say that the specimen although very small is distinct enough



for observation, when magnified. I understand it is from near the entrance to Tidal Inlet, Glacier bay, at an altitude of three or four hundred feet on the mountain side where he found a small depression a few inches deep and perhaps two feet across partially filled with water. The surface of the stones which projected through the water looked as though they were pointed with vermilion. He brought a stone home with him and submitted it to Dr. F. H. Herrick of Western Reserve Uni-

*A limited number of slides illustrative of this article can be had of the author at 50 cents each.

versity, Cleveland, Ohio, who said it was an almost pure culture of *Hœmatococcus*. He says that red snow was seen in several places but no specimen was brought back, evidently not referring to the vermilion colored stones. But this was colored with red snow also, brighter in color, but the microscope shows it plainly. The water was just above freezing, perhaps 33° Fahrenheit.

This gives me an opportunity to say something on the subject of red snow. This remarkable phenomenon which is known under this name has been a subject of very extensive investigation, and it is well known to be the result of enormous development of a microscopic organism which is called *Protococcus*, *Chlamidococcus* or *Hœmatococcus*. Dr. Henfrey says he is "induced to believe that more than one form is comprehended at present under the name of *Protococcus* or *Hœmatococcus nivalis*, for in a specimen of red snow (for which we are indebted to the kindness of Mr. R. Brown) appears to belong to the same genus as *Palmella cruenta*" as first indicated by Mr. Brown, and confirmed by Sir W. J. Hooker. Dr. Greville's figure of the Scotch plant closely resembles this; but the continental plants described by Mr. Shuttleworth and others, would seem congeneric with *Protococcus*, *Chlamidococcus* (*Chlamidococcus* Braun, *Chlamidococcus* Ehr.), since they produce negative zoospores, the forms which Mr. Shuttleworth described as distinct infusoria as species of astasia. Nearly connected with this continental snow-plant; if not identical, is the *Protococcus pluvialis*, described so elaborately by Dr. Cohn, which moreover appears synonymous with *Discerœa purpurea* of Morren. This was in 1856. In 1892 I published the examination made by means of the microscope of the specimens of the Geological survey of California. Therein is detailed, at page 24 the gathering of "red snow, from Lassen's peak, at about 10,000 to 11,000 feet, 1863. This gathering reached me dried upon

paper. It is one of the forms which have been described under the names of red snow or *Protococcus* or *Hœmatococcus nivalis.* It is one of the minute forms which have been described under *Palmellea* or *Euglena* and are but the motile conditions of highly organized algæ, Lichens or other plants." Vast changes have taken place since 1856, the publishing of Dr. Henfrey's Micrographic Dictionary. The works of C. Swendener in 1860-68 wherein he shows that Lichens are but the forms of plants which are made up of Algæ and Fungi, so that Lichens, although seemingly distinct plants, and placed in a kingdom by themselves, are compound and have no kingdom at all. But this is not all for the *Protococcus* or *Hœmatococcus* is found to be a fungus itself. And even the spores or seeds if a *Hœmatococcus* are seen to give forth Algæ and thus a Lichen. This was long disputed and Swendener's facts doubted, but now they are accepted by all of the modern naturalists who have studied them and the growth of Lichens themselves. From this there comes a process of thinking of what is a species in these plants and are they plants at all. For seeming animals which are active and take in food and assimilate it, grows to be plants. And *Hœmatococcus* is one of them. So that we cannot distinguish by any means we have species of an animal from another of vegetable.

Hœmatococcus or red snow, of which the specimen from Tidal Inlet is one, may be described as a stiff gelatinous mass, which is white and transparent, with diffused through it, certain spheres. These spheres in their turn are surrounded by a thick almost woody cell-wall, which is more transparent than the gelatinous mass. Within these cells are granular red or green coloring matter. So that the whole looks like and was thought formerly to be blood. Blood showers are commonly spoken of by the older inhabitants and even by some of the modern ones. We even now occasionally hear of bloody rain, which is

only the *Hæmatococcus* which appears after a shower of water on the soil, on the brooks, by the wayside. *Hæmatccoccus,* or rather when it is green *Protococcus*, is extremely common on the water of stagnant marshes. Sometimes it is red and then the *Hæmatococcus* appears. When this water dries up, or rather when the land is dryer, these may develop into Lichens or the green on barks and fences. For these are but the same, Algæ when in water, Lichens when on dry land. *Protococcus* of a green color when at the ordinary temperature and Red Snow when the temperature is freezing. To see the Red Snow understandingly it is necessary to magnify it four or five hundred diameters. That is to say one hundred and sixty thousand or two hundred and fifty thousand times, and this can be done by using an objective of ¼ or 1-5 inch and what is known as a B, or second eyepiece or ocular. A wide angled lens is not necessary, but certain minute markings can be made out with such a glass which are invisible to an ordinary lens.

There were seen in the specimen from Tidal Inlet besides the red spheres, which constitute the Red Snow itself, an individual of a stylospore, or stalked spore, of a Coniomycetal Fungus, showing that it lived at that temperature; i. e. about 33° Farh., and there are also common green Confervoid Algæ, belonging to the gems Oscillatoriaceæ, which are long, filaments with close transverse striae. When living they continually roll about and in this way move like animals.

In 1858 Mr. George Gibbs, Geologist of the North West Boundary Survey found at an altitude of 6500 feet on the Cascade Mountains the Red Snow. He disgusted the man who first brought it to him by eating it and examined by means of a lens, showed it to consist of "tadpole shaped bodies with rounded heads and alternated tails, perhaps two lines in length." In conclusion he says that he believes that is the first notice of the occurrence of

Red Snow within the territories of the United States.

Prof. Brewer informed me that he found Red Snow commonly on the perpetual snows of the Sierra Nevada. For many years before this I had seen this remarkable appearance, not however, upon snow, but upon the surface of water of ditches and marshes all around New York, This investigation has been treated of in a note on Colored Rain by the present writer in the proceedings of the New York Lyceum of Natural History, Vol. 1, page 272, October 23, 1871. I do not think the Red Snow occurs in a equable climate like that of San Francisco, or in the hot sun of New York, nor do I know of its occuring in the hotter climates of the South.

Diatoms of the Connecticut Shore.—VI.

By Wm. A. Terry,

Bristol, Conn.

In a former article of this series, October 1892, describing the diatoms of the Quinnipiac marshes, I mentioned a *Pleurosigma* which resembled closely *Colletonema eximium*; but which did not appear to be a *Colletonema* when I found it, but was an independent and rapid traveller like other *Pleurosigma*. The same form was abundant in the fossil deposit taken from Davis' pit, and the living specimens were found in ditches and pond holes in the adjoining marsh. They were very plentiful in the gathering and very active, perhaps more so than other kinds with them which were mostly *Navicula*.

With the material sent to M. Tempere from Davis' pit, I sent also a slide of these diatoms and called his attention to them. He replied that they were *Colletonema eximium*. In "*Le Diatomiste*" for December 1893, is an illustration of what appears to be the same diatom, Plate III, Fig 3. Prof. Cleve calls it "*Gyrosigma temperei*, Cl. n. sp," and says; "Valve linear straight, rounded

at extremities, length 0.14 mm., width .014 mm., median line straight, curved at extremities, longitudinal striæ 30, transverse 27 in 0.01 mm. Habitat, salt waters, Connecticut."

The Connecticut form is about three times the length of "*eximium*" as given in Brit. Diatoms, and has finer striæ; and as it does not grow in tubes but is a free traveller, I have always thought since my first discovery of it more than three years ago that it required a special name; but the propriety of calling it "*Gyrosigma*" may be questioned.

Prof. Cleve proposes to divide the genus *Pleurosigma* into two, retaining this name for those with oblique striæ, and adopting *Gyrosigma* for those with straight. This name was given by Hassell, and was rejected by Prof. Smith, on account of the "alliterative blunder," De Brebisson also comments upon it with disfavor.

Plate III also shows in Figure 5, "*Navicula theta*, Cl. n. sp." which he credits to Oregon; fresh water fossil. This form is also very abundant in the Quinnipiac fossil material, with many other related varieties, some of which were rather doubtfully referred to *Navicula pusilla*, by Prof. Kain to whom I sent a slide. I think that several of these varieties are yet undescribed any further then in general terms in my former article; they were not included in the material I sent to Europe which came from Davis' pit; these varieties were in material from Share's pit, about two miles farther North. Three of these related varieties are very numerous in this material; they vary greatly in size and considerably in outline and also in fineness of striation; the larger variety is proportionally more elongated than the others and has finer and less distinctly marked striæ. Cleve's figure is an accurate representation of one of the smaller of these forms, excepting that it does not show the striæ so distinctly moniliform; the larger is more then three times

the linear dimensions of *N. pusilla* as shown in Smith's Brit. Diatoms, Pl. XVII, Fig. 145. Further examination of the Quinnipiac material shows many other species of which I can find no description in any publication accessible to me; many of these occur sparingly at other places along the Connecticut shore.

I had intended to devote considerable time early in the season of 1893, to a more thorough examination of the deposit at Leete's Island and of the neighboring waters; but the derangement of business caused by the World's Fair prevented, and I did not get at work there until the latter part of August, just before the great storm of that month. This was an unusual storm for this locality, and stirred up the usually comparatively quiet waters of the sound to an uncommon extent. Standing upon the point between Leete's Bay and Little Harbor, the dash of the breakers upon the outermost of the Thimble Islands was an interesting sight. On one of these islands was a seaside cottage surrounded by a grove of trees; the spray was sometimes driven fifty feet above the tops of the highest trees, and descended in a blinding cloud which would blot the entire Island from sight. The salt spray was carried inland and killed the leaves of the trees for a distance of two miles from the shore in some places, the fruit trees near the shore being entirely denuded of foliage and afterwards starting into flower. When I called on my friend Mr. R. C. Leete in October, his orchard of apple and plum trees was in full flower.

Assisted by Mr. Irving Leete I made soundings in Leete's Bay, in Great Harbor, and both sides the reef that extends westward from Sachem's Head, and outside the reef some miles into Long Island Sound, and westward toward Thimble Islands past the buoy that marks the channel. In all these places I found a soft bottom excepting in the outer soundings in the deep water of the sound. I had intended to explore the shallow waters

near the shore throughout this whole region, but judged that the disturbance caused by the storm had so mixed things that the normal condition could not be well ascertained.

A superficial examination of the material procured by these soundings shows the general resemblance that I had anticipated; a special feature is the great preponderance of *Coscinodiscus* from large to minute varieties; *Actinoptychus* is abundant, and *Pyxilla baltica* and *Pyx. dubia* especially plentiful; *Rhizosolenia styliformis* and *Biddulphia aurita* are also abundant with *Pleurosigma affine, Melosira sculpta* and *Rhabdonema adriaticum. Navicula lyra* appears in many varieties and *Triceratium favus, Eupodiscus argus, Raphoneis amphiceros* and *R. gemmifera, Surirella fastuosa, Pleurosigma balticum, P. decorum, P. wansbeeckii* and *P. minutum* are common. Very large specimens of *Navicula latissima* occur but are rare. Near the shores of the bay *Auliscus* and *Actinocyclus* appear in variety some of which I cannot determine, and a large *Navicula* with striation something like *N. latissima* but rather more distinct, and with a broad elliptical outline without the produced extremities. I have found this form in the fossil deposit here, and in recent mud from the Thames river near Norwich Conn. I do not think it has ever before been noticed.

Foraminifera of many genera abound, not only in soundings but also in the deposits in the creeks, ditches and marshes; at the upper end of Leete's creek at the limit of tidal flow I found but few diatoms, and those mostly small fresh water varieties; but foraminifera were abundant, of many species new to me; their shells did not appear to be calcareous but showed a mosaic structure with markings of a somewhat regular irregularity very similar to the shells of certain rhizopods, *Nebela collaris* and *Difflugia spiralis*.

Polycystina are common in soundings and deposits. I

have sometimes noticed living specimens of large size and peculiar structure, whose make-up caused me to suspect that certain forms illustrated in the books and classed in different genera were merely different parts of the same organism, but circumstances prevented the thorough investigation necessary to demonstrate the facts, and I therefore mention it merely as a suspicion. At the eastern end of Leete's Bay is "Shell Beach" extending from the main land to Leete's Island, and crossing the ancient channel which once surrounded the Island; this beach has a local celebrity on account of the number and variety of the shells cast up by the tide; among these are many species of small round clams, one of these averages about the size of a common cherry stone. A larger species has a bright pink color and very thin shell. The razor shell or Solen, *E. ensis* is numerous in all sizes, and *Fulgar carica* and *F. canaliculatus* are plentiful, *Murex*, *Helix*, *Helicina*, *Zonites*, *Nassa* and other genera with *Pleurotoma minima* and *Littorina obtusata* abound. After the storm, living specimens which had the form and outline of *Leda sowerbyana*, but which did not gap at extremities and had the polished surface of *Leda corpulonta* were left upon the beach. The row of minute sharp pointed teeth possessed by these shells is a peculiar feature of their structure. Near the point of the island and on the north shore of Great Harbor, I picked up two or three dozen of the rare shells of *Pandora ceylonica*.

Shell Beach is being slowly driven inland by the sea; its recession is laying bare deposits laid down many years ago, the diatoms of these deposits are much like those of the soundings. I find them difficult to clean on account of the quantity of minute scales of mica they contain, which are almost impossible to separate by ordinary methods. I have made a few slides by separating the heavier discoid forms by the process first pointed out by Christopher Johnston, M. D., in *The Lens* of No-

vember, 1873. In this process the diatoms and water are poured from one vessel into another; the first vessel is rinsed with water to wash out fragments and scales, then the diatoms adhering to the glass are brushed off with a small clean brush and decanted, this is repeated until enough are obtained comparatively clean. This process will only separate certain forms, chiefly discoid; although *Triceratium favus* and *Navicula lyra* will also go with them to some extent, but many species cannot be cleaned in this manner, and it will not give a just representation of the species to be found in a deposit.

I find in the Shell Beach deposit:

Actinoptychus undulatus, E.
" areolatus, E.
Actinocyclus barkleyi.
" ehrenbergii, Ralfs.
" crassus, Sm.
" subtilis, Cleve.
Amphiprora lepidoptera, Greg.
" conspicua, Grev.
Amphora ovalis, K.
Amphitetras antediluviana, E.
Auliscus caelatus, Bail.
" macraeanus, Grev.
" pruinosus, Bail.
" radiatus, Bail. (rare).
" sculptus, Ralfs.
Biddulphia aurita, Breb.
" laevis, E.
" pulchella, Gray.
" rhombus, Sm.
" turgidus, E.
Campylodiscus echeneis, E.
Cocconeis scutellum, E.
Coscinodiscus apiculatus, E.
" oculus iridis, E.
" gigas, E.
" radiatus, E.
" excentricus, E.
" marginatus, E.
" subtilis, E.
Cyclotella striata, Grun.
Cymatopleura marina, Lewis.
Epithemia musculus, Kg
Eupodiscus argus, E.
Isthmia nervosa, Sm.
Melosira arenaria, Moore.
" borreri, Grev.
" sculpta, E.
Navicula lyra, E. var.
" didyma, Kg.
" elliptica, Kg.
" smithii, Breb.
" maculata, Grev.
" humerosa, Breb.
" latissima, Greg.
" californica, Grev.
" prætexta, E.
" theta, Cl. n. sp.
" hennedyi, Sm.
Nitzschia circumsuta, Grun.
Plagiogramma validum, Grev.
Pleurosigma affine, Grun.
" balticum, Sm.
" decorum, Sm.
" minutum, Grun.
Pyxilla baltica, Grun.
" dubia, Grun.
Podocystis americana, Bail.
Rhabdonema adriaticum, K.
Scoliopleura tumida, Breb.
Stephanopyxis turris, E.
Surirella fastuosa, E.
" striatula, Turpin.

Stauroneis aspera, Kg.
Raphoneis gemmifera, E.
" amphiceros, E.
Hyalodiscus radiatus, E.
Rhizosolenia styliformis, Bright.
Triceratium favus, E.

The above list does not include the whole number of species found in this deposit, possibly not more than half of them; but enough are given to show the general character of the deposit. I have not mentioned quite a number of the larger forms because I felt uncertain as to their proper determination; and multitudes of the minute kinds are omitted for the same reason. The Connecticut shore material is remarkable for the large amount of very minute forms it contains. I had hoped to investigate the deposit thrown up by the pressure of the R. R., embankment crossing Leete's Creek, from top to bottom during the dry weather but did not make it out. We selected a place where the earth had been thrown up and cracked open exposing a stratum about seven feet below the original surface, here we dug down five feet and procured earth from at least twelve feet below the top. Slides made from this material show beautiful specimens of *Surirella febigerii*, *Tropidoneis seriata* in great abundance, *Pleurosigma balticum*, large *Coscinodiscus*, large *Synedra*, *Actinoptichus undulatus*, *Scoliopleura* and *Navicula* all abundant; and small kinds down to very minute in about equal proportion. I sent a slide containing the medium density to Dr. D. B. Ward. He reports: " In the 12 foot deposit slide I found *Navicula aspera*, *didyma*, *marina*, *fischeri*, *elliptica*, *elliptica* var., *oblongella*, *peregrina*, *digitoradiata*, *yarrensis*, *lyra*, *interrupta*, *smithii*, *formosa*; *Stauroneis salina*; *Amphora proteus*; *Pleurosigma affine* and *balticum*; *Tropidoneis seriata*; *Rhabdonema adriaticum*; *Scoliopleura tumida*; *Actinoptychus undulatus*; *Nitzschia marginulata*, *sigma* var.; *Surirella febigerii*; and a small var. of *fastuosa*; *Paralia sulcata*; *Coscinodiscus oculis iridis*, *radiatus*, *excentricus*, *apiculatus* (?); *Synedra fulgens*; *Biddulphia aurita* and *rhombus*."

Near the R. R. station at Leete's Island is a small fresh-water pond used as an ice pond, whose bed was once a salt marsh; in this pond I found abundance of living specimens of a small *Pleurosigma* I had never met with before, *Pleurosigma subsalinum.* The valves of this species are so exceedingly light and delicate as to be nearly invisible, and I might not have seen it if I had not found it living; the median line has a peculiar bend resembling a sickle. Dr. Ward writes: "You are undoubtedly right in naming the *Pleurosigma P. subsalinum.*" It belongs to Peragallo's eighth group, but the markings are finer than those he gives, I had difficulty in resolving the longitudinal lines with a good homo immersion 1-12 (Reichart) by central light, though they came out well by oblique illumination." He adds, "the Beatties Pond slide is one of the most interesting I have seen for a long time, I looked it over last night and made a list of the forms I found. There are a good many others I could not name off hand, and several which are new to me, I found dozens if not hundreds of *Navicula peripunctata*, Brun. (Esp. Nuov. Pl. XVI, Fig. 11). Brun gives Crane Pond as the source of his original specimen. I found *Navicula peripunctata, viridis, peregrina, smithii, interrupta, limosa, astuarii* (Cleve), *crassinervia, nobilis, delawarensis, formosa, gibba, producta, elegans, major, borealis, ligumen, etc; Nitzschia tryblionella,* (2 var.) *plana, scalaris* and *obtusa; Surirella gracilis; S.* (*tenera* var?) *terryana: Achnanthes subsessilis, Coconema scutellum; Pleurosigma affine; Cyclotella striata: Strauroneis gracilis; Eunotia major* and *arcus; Gomphonema accuminatum; Raphoneis amphiceros; Amphiprora lepidoptera; Van heurckia rhomboides* var.; *amphipleuroides; Tabellaria fenestrata; Synedra pulchella; Coscinodiscus excentricus; Actinoptychus undulatus; Mastogloia angulata.*"

I have the Crane Pond material to which Brun credits his specimen, but the Beattie's Pond material has hun-

dreds where the other has one. To Dr. Ward's list above I would add "*Navicula theta*, Cl. n. sp." *Pleurosigma subsalinum* and *simile*, "*Gyrosigma tempereii*" Cl. n. sp. I explored Pine Orchard Creek but found only the usual kinds. *Pleurosigma americanum* is found in all the tidal creeks I have so far examined. The rail road crosses the head waters of this creek and the pressure of the new enbankment has thrown up the earth on each side but has not exposed the deposit. I dug down in one of the largest cracks and procured earth from about seven feet below the surface, but found the deposit to consist entirely of fresh water diatoms. *Stauroneis baileyi*, *Pinnularia lata* and *Navicula americana* were characteristic forms.

Rodger's Creek near Leete's Island Depot is rich in *Pleurosigma*, containing nearly all the common kinds, with abundant *Nitzschia* and *Synedra* and very large *Melosira borreri*. I took material from the numerous pond holes in the marsh, but have not yet found anything new in them and have given them only slight examination. The deposit underlying the marsh contains similar forms to the other Leete's Island deposits, but has not yet been carefully studied. From the shore to the Depot at Sachem's Head, a long cove extends about a mile in length, and is called "Long Cove." This contains *Pleurosigma terryanum*, *Navicula maculata* and *N. permagna*. The pond holes in the marsh at Sachem's Head are very rich in *Pleurosigma balticum* var. *maxime;* they are near the salt water, and I find that shallow salt pools often contain *P. balticum*, those farther back and more brackish are likely to have *P. elongatum*, while those very nearly fresh have sometimes *terryanum*.

Here I would like to give a caution to collectors; the material from such pond holes and tide pools has usually very little sand, but is chiefly composed of organic matter; this becomes quite firm before it is anywhere near

dry, and afterward shrinks greatly if allowed to dry completely. This shrinkage crushes the diatoms into fragments, particularly if such large and fragile species as *Pleurosigma balticum* and *P. terryanum* are concerned. In soundings and in fossil deposits, the large amount of fine sand protects the diatoms to some extent, so that they are not so certain to suffer in drying, but fossil material should be handled with great care. I frequently see it sent out pulverized, as if the senders imagined that such small forms could not be broken. Some recommend breaking down refractory material by soaking in water then freezing and thawing, this is ingenious but slow; why not use a hammer? It would be quicker and not much more destructive. I have sometimes found that in digging fossil earth the pressure of the spade in lifting out blocks of material would compress the earth sufficiently to crush millions of the best diatoms it contained.

On Cements for the Microscope.

By ARTHUR M. EDWARDS, M. D.,

NEWARK N. J.

With the danger of trespassing on the ground occupied by other observers and more thoroughly worked over by them, I nevertheless venture to write what I have arrived at in the way of using cements for the preparation of microscopic objects. I have used them for over forty years and have always turned back to English gold-size, as being the best to use in all cases. It is tough and dries readily enough, being about a day in drying.

Now what I use is this: Some modification of Shadboldt's turn-table is necessary. My friend Col. Kinne of San Francisco, made the first and on all accounts best, I believe. But to Mr. George Wale is due the iron stand. I remember, over twenty years ago, when he brought it

out. It was marvelous, and at the meeting of the old American Microscopical Society, which was named, not by me but by others "of the city of New York" as if that was necessary. I named it the American Microscopical Society when there was only one other Microscopical Society in existence, the Royal as it was afterwards called, of London. I say my friend Mr. George Wale brought out the iron stand for the Shadboldt's turn-table, and many wonderful contrivances he invented there. Witness the microscope stand with an entirely new form of limb, which is figured and described in the seventh edition of Dallinger's Carpenter on the Microscope. The slide is placed on the turn-table and centred. Then it is turned around slowly by means on the forefinger of the left hand whilst the brush, (I use a very small brush), with cement in it is brought down in the right hand to form a ring of cement. This must not be too thick at first but allowed to dry thoroughly. Next day when it is dry, (it will dry quicker in summer than in winter), another coat is put on and three coats are enough unless a thick cell is wanted, when another cement bearing a solid in it, as white zinc or umber or something else like that is used.

When the cell is thoroughly dry, and generally this takes a week at the shortest even in summer, the slide is put on the turn-table and a thin layer of gold-size is put upon it. Now the object, the diatom, alga, or animal specimen, is put in the cell, the watery preservative put in and the cover, held with a fine forceps, lowered one side at a time, so as to sweep a wave of preservative before it, into place. The edge of the cover is wet with the new gold-size and the cover itself pressed down into place. The extra preservative washes out and is absorbed with an old handkerchief used lightly. When the cover, cell, and slide is quite dry, a ring of gold-size is now put upon it and it is set aside to dry. Two or three

coats of gold-size are then, in the same way, put on it. I do not think that more coatings of gold-size are necessary and the cell can be finished with a coating of a fancy cement if it is wished. Blue, red, white, yellow or black will do. But with slides for my own cabinet, I prefer gold-size alone. This is all, and the object will keep generally for years. When exposed to wide variations of temperature, as are common in this climate, no cement is always trusty. A majority of slides keep well but a minority spoil by leaking. I say the preservative must be watery, for strong alcohol or, what is worse, bisulphide of carbon or turpentine will dissolve the wall of the cell and the slide will inevitably leak sooner or later.

Now I come to the use of other cements than gold-size for microscopic purposes, and first, asphaltum. Asphaltum dissolved in turpentine is used as a dryer by the painters, but asphaltum varnish contains more asphaltum than this. Trinidad asphaltum is about the only asphaltum in commerce, I believe. But it is cheap, and it makes a good durable cell. But it dissolves in weak alcohol and in some other preservatives and therefore cannot be used.

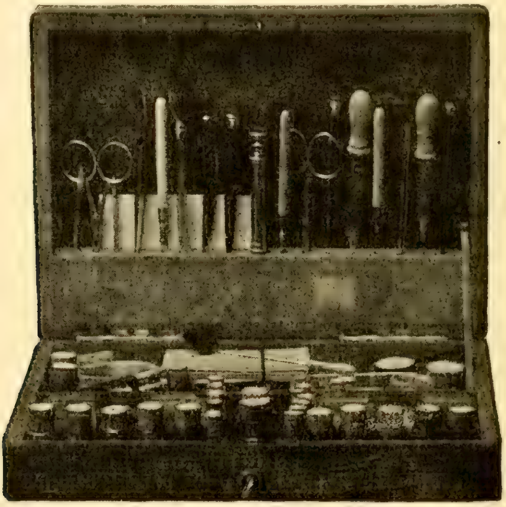
White zinc made up with drying oil, or white paint makes a pretty good cell; and red, vermillion, venitian red, and ochre, yellow ochre paints make pretty good cells also. But when gold-size is added to the paint, an excellent cell varnish results.

What I have found makes excellent varnishes both for cells and finishing are the different colored Star Enamels. They are fine in texture, dry quickly and evidently contain a varnish for they make a bright surface. I use them for cells constantly. White for cells and oak color or mahogany color for finishing. Bright red, green, blue, yellow, or black make excellent varnishes for microscopic use.

A Club Working Case.

By A. T. ELWELL,
COUNCIL BLUFFS, IOWA.

While the usefullness of portable microscopes, for certain work has been favorably mentioned in the Journals from time to time, the importance of a companion piece, in the way of a compact working case, has received little notice. Believing there is a field for such an accessory, and that others may be interested in that direction, I will refer to the one shown in the cut. In size it is seven



by nine inches, and less than two in depth. A slab of cork is attached to the lid holding in position a number of instruments, while compartments are provided for slides, cover-glasses, cells, and other material, leaving space for a number of bottles of stains, reagents, cement, and objects for examination and mounting. Intended as an auxiliary to the portable microscope, it should correspond in size with the case of that instrument, so that when strapped together for portability they will form a package of uniform proportions.

EDITORIAL.

The use of Formalin in Hardening and Fixing Tissues.—At the meeting of Jan. 9, 1894, of the Microscopical Society of this city, Dr. W. W. Alleger demonstrated a method of preparing tissues so as to show bacteria, the new feature of which consisted in the use of formalin as hereinafter described.

The tissue selected was from the lungs and liver of a guniea pig dead of anthrax. It was hardened by immersion for 48 hours in a 10 per cent solution of formalin, dehydrated in alcohol, cleared in chloroform and imbedded in paraffine. Sections were cut on a B. and L. Microtome, attached to the slide by means of gelatine fixative containing formalin, and after the fixative had thoroughly dried they were passed through turpentine to remove the paraffine, then into alcohol and finally into water. They were now stained for 5 minutes in an aniline water solution of gentian violet decolorized in a weak solution of iodine (Grams method) dehydrated in alcohol, cleared in cedar oil and mounted in balsam in xylol. A modification of the method mentioned consists in the use of aniline oil slightly tinted with eosine, in place of alcohol as a decolorizing agent. Thus treated the bacteria are stained a deep purple, the nuclei a light purple, the protoplasm a light rose and blood corpuscles brown, the result bring a beautifully stained and clearly differentiated mount.

Dr. Alleger claimed that tissues prepared in this way are superior to those hardened in alcohol in that there is much less shrinkage, and stated that while he was aware that bacteriologists generally do not look upon the paraffine method with favor he preferred it because of the freedom from the trouble of immerson fluids and for the reason that the tissue can be kept in blocks indefinitely and is always ready for cutting. He put most stress, however, upon his method of fixing the sections to the slide which is a modification of Dr. Gray's gelatine fixative method. To a dram of ½ per cent solution, he adds 4 or 5 drops of formalin. A drop of this mixture is placed upon the slide, the section laid upon it and gentle heat (not enough to melt the paraffine) applied. No matter how wrinkled the section it floats out flat and as soon as this occurs the excess of fixative is drained off and the adherent portion allowed to dry, when the

section will adhere firmly through turpentine, alcohol and aqueous dyes, staining is in no wise interfered with.

Dr. Alleger recently informed us that since the happy thought of using formalin to render gelatine fixative insoluble occurred to him he has discarded all other fixatives using this both for individual and class work and that during the entire work of the session just drawing to a close, neither he nor any member of his class has had a section thus fastened to the slide slip or become detached in the ordinary process of staining. Nothing less than a stream of water or other mechanical means being sufficient to loosen a section when thus (properly) fixed. This is a result he has been unable to secure in any other manner and is regarded as supplying a long felt want.

MICROSCOPICAL APPARATUS.

Price of *Zeiss's* Objectives. — The following announcement has just been received :

Having, through nearly ten years practice, gained a greater experience in the production of the apochromats, and having recently succeeded in materially simplifying the construction of these lenses, we have been permitted to make a *considerable reduction* in their prices.

LIST OF THE APOCHROMATIC OBJECTIVES.

	Numerical aperture.	Equivalent focus in mm.	Initial Magnification.	Price: Marcs.	
				former.	now.
Dry Series..........	0.30	24.0*	10.5	[140]	120
"	0.30	16.0	15.5	[100]	80
"	0.65	12.0*	21	[170]	140
"	0.65	8.0	31	[130]	100
"	0.95	6.0*	42	[220]	180†
"	0.95	4.0	63	[180]	140†
"	0.95	3.0	83	[200]	160†
Water Immersion.	1.25	2.5	100	[300]	250†
Homogeneous Im.	1.30	3.0	83	[400]	300
"	1.30	2.0	125	[400]	300
"	1.30	1.5	167	[450]	350
"	1.40	3.0	83	[500]	400
"	1.40	2.0	125	[500]	400

*The three objectives 24 mm, 12 mm and 6 mm of the dry series are constructed exclusively for the 10-inch tube ; all the others are adjusted either for Continental or English tube. † With correction collar.

In the foregoing table we give, at the side of the previous quotations [in brackets], the prices at which the apochromats will be sold henceforth.

At the same time we beg to announce a modification concerning the achromatic objective 1-12 (Homogeneous Immersion). This objective which we used to construct either with aperture 1.20 or 1.30, will in future be only supplied with one aperture—1.25 in the mimimum—at the price of M. 160.

CARL ZEISS,
JENA, Feb. 15, 1894. *Optische Werkstatte.*

Substage Attachment.—The substage attachment described by G. W. Brown, Jr., in the December Journal, is made by Joseph Zentmayer of Philadelphia, and is sold in connection with the Abbe condenser and iris diaphragm for about $30. For further particulars apply to Joseph Zentmayer, 209 South 11th Street, Philadelphia.

MICROSCOPICAL MANIPULATION.

Transmitting Slides.—The tin-boxes in which the typewriter ribbons are packed are 3½ long by 1½ inches wide and make most excellent boxes in which to transmit slides. There is just room enough to insert the slides wrapped in cotton or in blotting paper and no damage can be done by the mailing or by the pressure to which packages are subjected in mail bags. This is of especial importance when slides are sent across the ocean.

To Centre a Slide.—A "village druggist" sends us the following:

Put into a watch glass a pinch of prepared chalk and enough methylated spirit to make a thin white-wash. Mix, and with a camel hair pencil smear over the central part of both surfaces of the slide. Stand on edge to dry. Now make a tool something like a carpenters marking gauge in miniature. Just a little rod of wood about 1·5 inch in diameter and 3 inches long with a little block slid and fixed on it. With a triangular needle (which every one should possess), drill two holes in the rod; one, say 6-10 inch from one face of the block: the other 1 4-10 inch from the other face. Into these holes fix little pegs of wood to pro-

ject about 1-10 inch; these make the mark. By this time the slide or slides—the writer always does a packet at a time—will have become dry. Now work the gauge round the four sides of the slide ; the result will be a small square absolutely central ; also a maltese cross, the arms of which serve admirably as a guide for placing the cover. Pack the slides close together in a box alwavs with the marked face in one direction. Before using a slide just quietly wipe off the chalk from the blank surface.

To Cut Hard Chitinous Objects.—Minchin, while at the Naples Laboratory, succeeded in cutting sections of eggs containing yolk, such as cephalopods and other hard objects as follows :

Make two solutions in ether (1) of celloidin or collodium (2) of gum mastix, having added a small quantity of absolute alcohol. The celloidin should be as thick as possible and the mastix as thick as syrup. Mix equal quantities of the two solutions.

When a hard object is to be cut, thin some of the mixture with ether mixed with a little absolute alcohol until the mixture is quite fluid. The object to be cut being embedded in paraffin and placed in the well of the microtome, paint the surface of the paraffin block with the thin mastix-celloidin solution by using a soft paint brush. The solution sinks into the paraffin, the ether evaporates leaving the mixtures near the surface of the block. Wipe off the excess and polish the surface by rubbing with the finger. Now cut a section and behold the matter held together in a strip iustead of crumpled and scattered in fragments. Repeat the process of coating with mastix-celloidin for each section cut and whole series of uninjured sections can be cut through eggs containing yolk, through the thorax of an insect and many other refractory specimens.

To Mount Protozoa.—A drop of water containing the animals is placed in a slide and covered by a cover-glass supported at its four corners by wax feet. The drop should be free from debris of all kinds. The wax feet should be high enough to keep the organisms from being squeezed, but low enough to keep them from rapid motion—the slip should just not touch them. A drop of Hermann's or of Fleming's solution should then be placed at the side of the coverslip and cautiously drawn through by blotting paper or filter paper being placed at the

opposite side of the coverslip. Be careful that these living and freely swimming animals are not swept out from under the cover glass by the current before the reagent has reached them. This may be done by drawing the reagent half through and then putting another drop on the other side of the cover-glass and drawing it back again. When the reagent reaches them it both kills them and sticks them to the slide, after which the rapid currents will not move them. After the reagent has acted a few minutes, it is washed out with water, drawn through by means of filter paper. When well washed run in the stain (alum carmine or carm-alum). Let this stand an hour in a damp chamber (to prevent evaporation) and then wash with water, bring up through the alcohols into oil of cloves and finally into balsam. This method preserves cilia, undulating membranes, etc., and gives good results for the nucleus and internal protoplasmic scructures.—*E. A. Minchin in Natural Science.*

MEDICAL MICROSCOPY.

Cauterizing wounds.—A. E. Baldwin takes a 5 inch sunglass, brings it to a ¼ inch white focus upon the wound. The parts to be burned are first shaved and cleansed with a 1-10,000 solution of corrosive sublimate. The application is for 2 to 2½ minutes. There is no bleeding. Its use upon boils is very satisfactory and completely aborts them. It cures hemorrhages and glanders.

Amoebæ were found by Posner (*Berlin. Klin. Woch.*) to be the cause of a case of haematuria. The amœbæ are described as masses of granular protoplasm ten times as large as colorless blood corpuscles.

BACTERIOLOGY.

Gunther's Bacteriology.--Dr. Carl Gunther has just published at Leipzig, a new edition of his text book on bacteriology, the first edition having appeared in 1890, the second in 1893 and the third in 1894.

In the description of the cholera comma bacillus has been added a large amount of fresh material. Methods of isolation

are given. A newly discovered bacillus the *vibrio berolinensis* seems closely allied to the cholera bacillus, it being distinguishable only by the appearance of the colonies on gelatine plates. Even then, the finer granulation of the contents and the less irregular contour of the colonies are differences so slight as to occasion uncertainty at times. The *vibrio aquatilis* recently isolated from water by Gunther, is described and figured.

Bacterium Coli Commune is considered by Ekehorn a cause of appendicitis.

DRUG AND FOOD ADULTERATION.

Dilution with tumeric.—Hans M. Wilder has found that powdered rhubarb and yellow mustard are adulterated with tumeric. He detects the adulteration easily. Mix a little of the suspected powder with any volatile oil such as sassafras, fennel, anise, etc. Any of these oils dissolve the coloring matter of tumeric but do not affect mustard or rhubarb. The smallest speck of tumeric examined under the microscope will then show a yellow zone about it. The absence of tumeric is demonstrated by the microscopic field remaining colorless.

MICROSCOPICAL NOTES.

Directory.—The attention of those interested is called to an International Zoologist's Directory, soon to be issued by R. Friedlander & Son of Berlin. Its object is to bring into closer correspondence those who are working in zoology. If you wish your name to be inserted send it together with your address and mention your official position if you have one. It will include collectors, draughtsmen, preparators, modellers and dealers. Address R. Friedlander & Sohn, Carl-Strasse 11, Berlin, N. W.

Ice Dangerous.—However pure in appearance, ice may contain disease germs capable of development when taken into the system or brought into contact with food or drink. The evils attributed to the temperature of ice may be due to the germs.

Herbert Osborn's paper in the February number is spoken highly of by several subscribers who ask to have the series continued.

DIATOMS.

How to find Diatomaceous Earth.—Mr. W. A. T·rry of Bristol, Conn., sends us the following:

It is asked how we know when we have found Diatomaceous Earth? It is of course impossible to be absolutely certain until the find has been examined under the microscope, still a collector of experience and good judgment will make few mistakes. The fresh water deposits are a'most invariably covered by a layer of muck or peat. A light colored stratum below muck will be either *lime*, *clay*, or *diatoms*, if it is not *sand*. Sometimes *all* mixed. If the material dries of a light gray or ash color and is also light in weight, it is almost certain to be rich in diatoms. If the deposit is heavy, it is mostly clay or exceedingly fine silicious dust; it is always well to examine all such deposits under the microscope, and, if not rich they may contain rare varieties.

The marine deposits underlying the peat of all salt marshes of the Atlantic coast always contain diatoms; in some localities these are comparatively rich, and frequently contain deep water forms. The diatoms are not equally distributed through these deposits, but are apt to lie in thin streaks or in pockets; and strata holding shallow water forms and those containing deep water kinds are sometimes in close proximity. Where ditches have been dug through these marshes for drainage, material containing very interesting varieties is frequently thrown up. Shallow water kinds usually predominate near the surface, and below are streaks of deep water forms alternating with those of brackish water down to a depth of over twenty feet in many places.

On the New England coast, it is not very unusual to find these deposits to be from thirty to fifty feet in thickness. Where the deposits were laid down by strong tidal currents, few diatoms will be found. They deposit in eddies and basins after the coarse sand has been left behind.

These hints will be sufficient to guide the efforts of persevering and intelligent investigators in search of Diatomaceous Earth; but they must understand that tiresome and disagreeable labor is inevitable if they would win success. At some future time I will give more detailed instructions.

MICROSCOPICAL SOCIETIES.

San Francisco, Cal., Geo. Otis Mitchell, Sec'y.

March 7, 1894.—Mr. K. H. Freund spoke upon Paul Ehrlich's method of staining the cell elements of the blood. He gave a brief and interesting resume of our knowledge of the blood from the time when Athanasius Kircher with his simple lens, magnifying thirty-two diameters, observed and mistook blood and pus corpuscles for worms. Later, in 1658, Swammerdam observed the corpuscles in the blood of the frog, and in 1661 the great Leeuwenhoek reported to the Royal Society his discovery of the red corpuscles in the blood of man. Ever since then investigations into the nature and constituents of blood have held the attention of numerous and enthusiastic workers. Apart from mere histological interest it is evident that many questions involving life and health can be answered only after a closer knowledge of the obscure phenomena manifested in this vital fluid, and their significance. As Goethe said, " Blood is a very peculiar juice." Chemistry has done its share in increasing our knowledge of the subject, but the greater part of the work lies within the province of the microscope. Rapidly enumerating those whose investigations have thrown the most light upon the subject, the author came to the new era inaugurated by Dr. Ehrlich in 1880, when he announced his investigations as to the " granular cell" by means of what he called his " tinctorial or color analysis." This method is based upon the micro-chemical behavior of the granulations of the protoplasm in certain groups of leucocytes, or white blood corpuscles, toward a number of analine colors, and these effects are so constant and so marked that his method has become an indispensable factor in the clinical examination of the blood. Not only have Ehrlich's researches into the nature of the granules occurring in the leucocytes become means of the greatest histological and eventual pathological value, but have also furnished the way to ascertain the general condition of the blood and to obtain an exquisite picture of its cellular elements. The granules in the leucocytes present remarkable differences in their staining properties, differences which have both a physiological and pathological significance. Upon this basis he distinguishes five varieties of what he calls " granules," designating them by the first five let-

ters of the Greek alphabet. The employment of Ehrlich's stains for clinical purposes is at present mostly confined to the differentation of the pathological conditions known as leucocytosis, leukæmia and anæmia. After discussing the nature of these pathological conditions Mr. Freund gave a practical demonstration of the employment of the stains, carefully detailing each step from the extraction of the drop of blood to the finished and mounted preparation. He spoke with enthusiasm of *gum thus*, the new mounting medium which Dr. Eisen presented to the notice of the society at the last meeting.

After the demonstration, under a dozen microscopes were displayed preparations rendering evident the nice differential stainings, for which Mr. Freund is so well known.

Washington, D. C., L. M. Mooers, Sec'y.

February 14, 1894.—The society at this meeting held the third of a series of working sessions, at which Dr. Richard Foster, of Howard University, exhibited a method of demonstrating vegetable histology. Especially in class work the detail of preparing should be easy and ready. The doctor showed how the various parts of the plant may be sectioned, stained and mounted ready for examination in but a few minutes. Instead of glass, slips of cardboard were used with a circular opening punched in the center. The section, after being stained in an alcoholic solution and pressed with blotting paper, is placed between two films of mica, put upon the slip and covered with gummed paper also punched to correspond witn its cardboard slip. When finished the mount is similar to Walter White's objects, and is not only neat but practically permanent. Mr. C. W. Smiley presented samples of diatomaceous earth from New Britian, Conn., containing the form *Stauroneis nuova britainica.* He hoped thatsome one would give a working session showing methods of cleaning these earths. Two applications for membership were received. The Society decided to hold its Annual Soiree in May.

Lincoln Microscope Club.—Roscoe Pound, Sec'y.

December 26, 1893.—No meeting was held on account of the meeting of the Nebraska Academy of Sciences at the same time.

Jauuary 30, 1894.—No business meeting was held. The eve-

ning was spent in examining slides and in conversation, etc.

February 27, 1894.—Officers for 1894 were elected as follows: President, E. T. Hartley; Vice President, Dr. H. B. Ward; Secretary, Roscoe Pound; Treasurer, J. S. Dales; Executive Committee, Dr. Bessey and Dr. Philbrick.

The time of meeting was changed from the last Tuesday to the last Wednesday of each month.

Among the exhibits, Dr. Bessey showed a piece of a leaf of Indian corn subjected to the action of chlorinated soda, used as a substitute for Potasium hypochloride which he was unable to obtain in the market. It cleaned very well and accomplished all that the latter would.

Ottumwa Microscopical Society, Ottumwa, Iowa.

September 21, 1893.—This was organized as a new society with Dr. L. J. Baker, President; Dr. J. F. Herrick, Secretary. Other members are Mr. E. M. B. Scott, Dr. J Williamson, Dr. S. A. Spilman, Dr. B. F. Hyatt, Dr. W. B. La Force.

NEW PUBLICATIONS.

The Microscope and Microscopical Methods. By Simon Henry Gage. Fifth edition, rewritten, pp. 165, 8°. Ithaca, N. Y., Comstock Publishing Co. 1894.

This edition has been greatly enlarged (nearly one half) by the elaboration of the matter in previous editions. A chapter has been added on photo-micrography and on photographing natural history objects in a horizontal position with a vertical camera. The number of figures has been increased to 103.

This volume constitutes Part I, on the Microscope and Histology. Part II, will give the application of the microscope to the study of Vertebrate Histology and will be published as soon as practicable.

This as previous editions appears with half the pages blank for annotation. This is unnecessary so far as the general reader is concerned but will be useful when the book is used in college classes. As we have before said, the book is the best we have seen for class instruction, and its references being mostly to American apparatus, we should prefer it to English works which refer to foreign apparatus.

ROBERT B. TOLLES.

THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. APRIL, 1894. NO. 4.

On the Means of Distinguishing Human Blood.

BY HENRY L. TOLMAN,

CHICAGO, ILL.

[Address before the Microscopical Section of the Chicago Academy of Sciences, January 12, 1894].

Although the question of distinguishing human blood from that of other animals may appear to be already decided, yet it is by no means settled, and in fact the study is yet in its infancy. There are so many factors involved, so many nicely balanced questions to be considered, that the subject is one of great difficulty. Much of the work that has already been done must be done over again, all of it must be carefully revised in the light of recent attainments in technique and objectives.

Let us then consider the subject theoretically and decide what steps should be taken if the examination were to be made for the first time. The blood is a fluid of complex chemical composition ; is it not possible to find some constituent in one kind, human blood, for example which is not found in other kinds, that of the lower animals? We know that man eats different food from other animals, that he lives differently, that he is protected by clothes and sheltered in a home, that he is able by his intellect to avoid dangers and in a measure to absolutely neutralize the law of natural selection and survival of the fittest. Now of course any long continued cause acting on an organism, will necessarily modify it to a greater or less extent, and it seems natural that the human organ-

ism, living a peculiar life, would not only necessarily appropriate the chemical constituents suitable to that life, but must appropriate them in a different proportion from any other animal. Unfortunately, the discoveries in this direction, though showing much to support this view, have not yet reached that degree of certainty, which make them reliable indications; whether this can be done, is and important question for the future to settle. In my own experience I have thought that the color of the human blood corpuscles was so different from those of the dog, as to afford a valuable means of discrimination between the two, but I am not prepared to say, that it can always be relied on. This also would depend on first the amount of hemoglobin present and second on the density or specific gravity of the blood corpuscle itself.

There are four tests for the presence of blood, but only one for the differentiation between human blood and that of other animals, namely, that of the measurement of the blood corpuscles. Of the chemical tests by guiacum and the formation of hematin crystals, it is not the place here to speak, partly because they belong to the domain of chemistry, partly because they only go to the length of declaring that a fluid is blood of some animal. The spectroscopic test has the same defect, though as the microscope is frequently employed, it properly comes within the range of subjects to come before a microscopical society. This means of identifying blood is entirely conclusive for the spectra of meth-hemoglobin, hemoglobin, oxyhemoglobin and reduced hematin; cannot be confounded by a practiced observer with the spectra of any other substances.

Now as to the measurement of the blood corpuscles, which is of course, entirely a microscopical matter. The corpuscles, are as is well known exceedingly minute, flattened, circular, bi-concave bodies. So numerous that there are from 4,000,000 to 5,000,000 in a cubic millime-

ter of blood; so soft that they are flattened, elongated, rolled up, or changed into various other shapes as they rush along through the capillaries, yet so elastic that they instantly resume their form when the pressure is removed. They are of the highest necessity in the human economy, both as oxygen carriers, and as forming material for conversion into muscle, and hence on principle would be least likely to suffer sudden changes in form or character. When the blood is fresh, a small portion of a drop can be taken up on a dissecting needle, the needle laid flat on the cover glass, and drawn across it, leaving a very thin even film behind. This in my experience is the best way of obtaining a single layer of corpuscles evenly spread out. When the blood is dried in a clot on a board or tool, or piece of cloth, the task is much more difficult. The clot must be moistened by some fluid and the character of that fluid and its specific gravity are of the highest importance. On principle it is undoubtedly best to use some fluid as nearly as possible in chemical composition to that of serum. Hence, solutions of mercuric bichloride or even glycerine and water would seem to be rather less suitable than those containing sodium chloride, sodic sulphate or albumen, as these substances are natural constituents of the serum.

The specific gravity of this macerating fluid should not be less than about 1.050 to 1.055 as experience has shown that the maceration of the corpuscles has a slight tendency to enlarge them. Even a small variation in the density of the fluid may exercise an appreciable influence. There seems to be less power in the corpuscle when out of the body to resist the influence of a strange fluid, than when it is subject to the control of what is often called the vital principle. Hence although the specific gravity of the serum alone is about 1.028, it is not safe to use a fluid of that specific gravity for softening up an old clot, but the density should be about 1.050—the specific grav-

ity of the whole blood, including fibrin and red corpuscles.

The question as to whether corpuscles when thus soaked will resume their normal size, and how it can be known that such is the fact, is a very important one but it seems to me it can be settled to a satisfactory degree, by a consideration of the physical principles involved. The exterior portion of the corpuscle, although not a separate skin or covering, but an integral part of the cell, acts as a dialyzing membrane and allows the passage of fluids through it only in proportion to their density. Hence by the selection of a fluid with a proper specific gravity, we can feel assured not only that the cell will refuse to absorb the fluid and swell indefinitely, but that it will only absorb an amount equivalent to that which it had in the body during life.

Now before we begin measurement of the corpuscles, let us consider the principles involved and the tools we are to use. There are, as is well known, differences between the measurements of human blood corpuscles, by different observers, dependent on various causes.

It is an interesting fact that the tendency of the later work has been to show that the earlier observers made the measurements too small. The great weight of authority now is in favor of the statement that the average size of a human blood corpuscle is 1-3200 of an inch. For a long time, opinions were divided between 1-3300 and 1-3250, then the latter figure was accepted with reservations, then they wavered between 1-3250 and 1-3200 and now there are even some observers whose experience has been that 1-3200 of an inch is rather below than above the limit. My own observations have led me to reach this conclusion. The late Dr. J. B. Treadwell in an elaborate series of researches published in the Cyclopaedia of Medical Sciences makes an average a little over 1-3200. The late Dr. Richardson, whose extensive measurements are well

known, made an average of 1-3224 by measuring from the outside of the dark border on one side to the inside of the dark border on the other. By making the proper correction, so as to include the dark border on both sides, which should be measured the average is 1-3173. Dr. Ewell from a measurement of numerous cases also reaches a similar conclusion. This concensus of opinion in favor of larger averages, is due I think to greater accuracy in the character of instruments employed, and also more particularly to a better knowledge of optics. A human blood corpuscle consists optically of two kinds of lenses, the centre a bi-concave, dissipating the light, and the ring around it, a double convex, concentrating the rays. Outside of this ring is a black border, which was supposed to be caused by the diffraction of light striking the edge of the corpuscle. More careful consideration of the subject, however, shows that this black border belongs to the corpuscle, and that its color is due to the total refraction of light at the edge. There is of course also diffraction, but a wide angle immersion objective which is indispensable for such work, will pick up these scattering rays, and by its accuracy of correction and absence of depth of focus, enable the careful observer to focus exactly on the edge, where the breadth of the diffracted cone is at a minimum. Of the two kinds of eye piece micrometers, I much prefer the filar with both cobwebs adjustable. On a large number of observations, perhaps, the glass eye-piece micrometer would be equally accurate, the errors balancing one another, and it is much easier, more rapid, and less fatiguing to use but the unit of measurement is much larger, with a 1-10 inch immersion objective, 12 inch tube, ¾ inch eye-piece, and collar-correction nearly closed, the value of one division of the eye piece micrometer is about 1-80,000 of an inch, which can be estimated to one half or 1-160,000 of an inch, while with a filar micrometer under similar conditions

except that the eye-piece is a 1 inch, the unit of measurement is easily reduced to 1-430,000 of an inch.

In the selection of the corpuscles to be measured, all which are round and regular in outline and double concave should be included. This excludes all that have not filled out, all that by the absorption of too much fluid, have become globular or bi-convex, and all the minute blood placques, at times so common, together with white corpuscles. As a practical way to eliminate any possible bias, it is sometimes well not to ascertain the value of a subdivision of the micrometer wheel, or of a division of the glass eye-piece micrometer, until after all the observations are made. The measurements can be taken in the units of the wheel or scale without knowing their value. For instance, if a single division of the micrometer wheel with a certain objective is equal to 1-960,000 of an inch, and if this fact were known, there might be an almost unconscious tendency to make the readings give an average above or below that figure, according as high or low average would favor the theory of the case for which the measurements were made.

Now comes the important question of what value are these measurements. Admitting for the purposes of the argument that the proper steps have been taken for macerating the clot of blood, that the proper fluid was used, and that the various instrumental errors have been eliminated, is the result conclusive? There is an unfortunate misconception, particularly by lawyers of the meaning of the word "scientific." It is very frequently employed as synonymous with the term "mathematically correct" or "universally correct." Any deduction, even if not universally true, is a scientific one, if concerned with a scientific matter, and made according to the rules of logic. A conclusion is erroneously supposed to be only scientific when there are no exceptions to it, but unfortunately especially in medicine there are few

conclusions that can be drawn, to which there are no limitations. As regards the measurement of blood corpuscles, no one will dogmatically assert that human corpuscles can never be mistaken for those of any other animal. But even if there must be some reservation made in this general conclusion, the statement is still a scientific one, irrespective of its universality. More than that it is of value in the trial of a case where the origin of a given specimen of blood is involved, if it can be shown that the exceptions to the general rule are few or unusual. All authorities agree that the average size of a dog's blood is smaller than that of a man, but it is not possible to say with mathematical certainty that a given specimen of blood is human. Suppose that an observer could determine, in all but one case in a thousand, whether blood was that of a man or dog, would it not be valuable evidence? Certainly, there is very little testimony ever given in a court of as high character as this, and the same principle applies if the observer could only determine ninety cases in a hundred, the percentage merely being different. The difficulty with the law in the use of scientific evidence is that it demands more than science can possibly give. It asks that all conclusions should be without exception, and yet there is scarcely a general proposition in all the range of medical science, except perhaps in anatomy and surgery, which can be stated without limitation. Yet, in my judgment, the determination of human blood, as against all other animals except perhaps a dog, can be made with such a degree of certainty, as to entitle the evidence of a well qualified expert on that subject, to much weight. The narrow rule that the conclusion can at most be, that the blood corpuscles in a given case are consistent with those of human blood, does not, it seems to me, finely state the case. The expert must of course state the limitations or exceptions, but he should also be allowed with

equal fairness to say, as far as he can how much allowance should be made for these exceptions,—what percentage of chance there may be that he is mistaken. Some incomplete tables show that the possibilities, that the corpuscles of a dog's blood would measure as much as those of a man, are about one in sixty-two. As against all other common domestic animals the probability of error is much less. The subject is an interesting one, it is much too large to be condensed into a small space. Much of the work already done is unreliable, and I hope that this paper will have the effect of calling the attention of microscopists again to the matter, and of doing something toward stimulating the present uncertainty, and giving some facts which can be accepted beyond question.

Formalin in Bacteriology, with More Especial Reference to its Action on the Bacillus of Diphtheria.

BY DR. W. W. ALLEGER,

WASHINGTON, D. C.

[Read before the Microscopical Society of Washington, Jan. 9, 1894.]

Formalin is the proprietary name of a 40 per cent solution of formic aldehyde recently put upon the market by Schering, of Germany. Formic aldehyde is a gas having the composition CH_2O. According to Roscoe and Schorlemmer it was first successfully prepared by Hofman in 1867 by passing the vapor of methyl alcohol (wood spirit), together with air, over ignited platinum wire ; it is also known as the oxide of methene and as the aldehyde and ketone of formic acid.

So far as I am aware formic aldehyde has been put to no practical use prior to the manufacture of the concentrated solution by Schering; and little is yet known of its properties aside from the information contained in the circular issued by him.

Early in October the American agents, (Schering and Glatz, of New York), kindly sent me a small vial of formalin, since when I have been testing it in various ways in the bacteriological laboratory at Howard University and have undertaken some experiments with it upon the bacillus of diphtheria. These have not as yet been completed, but so far as they have been carried are eminently satisfactory and seem to show that in formic aldehyde we have an agent which is superior in its germicidal action to corrosive sublimate in solutions of a strength which can be well tolerated and which so far as I can determine is not injurious. I have not yet had an opportunity to test it clinically in cases of diphtheria ; but if my expectations are realized it will prove to be a great boom to humanity, and it is with the hope that some of my hearers will test it in the sick room that these preliminary experiments are now brought to the attention of the society.

Diphtheria is now known to be primarily a local disease. The bacillus which stands in an etiological relation to it develops at the point of inoculation, usually the mucous membrane of the throat, and does not invade the blood or organs unless it be under very exceptional circumstances, the secondary or constitutional symptoms being due to the absorption of soluble toxalbumins produced by the bacilli and not to the presence of the germs themselves. It is manifest, therefore, that if we can destroy the germs or inhibit their development in *the early stage of the disease*, before sufficient of the toxalbumin has been produced and absorbed to destroy the patient, we shall be able to avert a fatal termination and to rob this justly much dreaded disease of its chief terrors.

This statement is by no means based wholly upon theoretical considerations for it has been proved by carefully conducted investigations that in cases treated locally by germicides the bacilli disappear from the throat

at a much earlier period than in cases not so treated—a fact of importance in determining the limits of the period of quarantine—and clinicians have shown that the mortality and duration of the disease have been favorably influenced by such treatment when commenced early and efficiently carried out. We are all familiar with the excellent results reported to us in this disease by the use of sprays of bichloride of mercury by our friend Dr. Robert Reyburn.

Bichloride is, however, a dangerously poisonous drug and moreover has the disadvantage of forming with albuminous materials an insoluble albuminate which would seem to limit its action to the more superficial layers of the false membrane, unless used in excessive quantity, while other commonly employed germicides are open to the objection that they cannot always be used in sufficient amount and concentration to be positively destructive to the bacilli without injury to the healthy tissues or harm to the system generally. Upon reading Mr. Schering's claims for formalin to the effect that this agent is as potent as bichloride as a germicide without being so toxic and acts equally well in albuminous as in non-albuminous media it at once suggested itself to my mind as a valuable agent in cases of diphtheria and I accordingly set about testing its effects upon the diphtheria bacillus.

I first covered the bottoms of some Petri (double) dishes to the depth of from 1-16 to $\frac{1}{8}$ of an inch with glycerine agar, inoculated them with the diphtheria bacillus by drawing a platinum needle, dipped in a recent pure culture, several times across the agar carrying the needle to the bottom of the plate. I now lightly sprayed the surface with formalin varying in strength from a 10 per cent solution to 1 part in 1,000 and placed all the cultures so treated together with two similar cultures without the use of the formalin spray in the incubator. At the end

of 24 hours an abundant characteristic growth of the diphtheria bacilli had developed in the two control dishes but no growth was visible in those treated with the formalin spray nor did any growth subsequently occur therein the plates being kept under observation for a month.

Next, plate cultures were made in the usual way by introducing the bacilli into glycerine agar made liquid by heat, pouring the melted medium thus inoculated into Petri dishes, and allowing it to solidify before using the spray. Two were now sprayed with a 1 per cent; two with a 1 to 1,000; two with a 1 to 2,000; and two with a 1 to 10,000 solution of formic aldehyde and placed in the incubator with two control plates. Characteristic colonies developed in the usual time in the control plates. In those sprayed with the 1 to 100 and 1 to 1,000 solution no colonies developed. In one of those sprayed with 1 to 2,000 a single colony developed and was found upon examination to be a mould fungus; the other plate remained sterile showing that while a 1 to 2,000 solution of formic aldehyde was sufficient to sterilize a culture of the diphtheria bacillus, it had no effect upon a mould fungus, a spore of which had accidentally fallen upon the agar in the preparation of the plate. In the two plates sprayed with 1 to 10,000 solution, a large number of colonies developed of various shapes and colors showing that bacteria from the air had been carried into the plates with the spray, which was not strong enough to inhibit their growth.

Although the amount of liquid carried to the plates by the spray was very small, yet I was not sure in the first series of experiments whether the entire thickness of the medium had been penetrated, or whether the formalin reached the bottom by means of the needle tracks; but in the second series it was clearly evident that a quantity of a solution so dilute as 1 to 2,000, scarcely sufficient

to moisten the surface, did penetrate to the bottom of a film of solid agar, thicker than the false membrane which usually forms in the throat in cases of diphtheria.

I next made several stick cultures in glycerine agar and dropped upon the surface of each tube, except the controls, 5 drops of a solution of formic aldehyde of the following strengths, 1 to 100, 1 to 200, 1 to 500, 1 to 1,000, 1 to 2,000, 1 to 5,000, 1 to 10,000 and 1 to 20,000. At the end of 48 hours the control tubes showed an abundant growth both upon the surface and along the entire needle track (1 inch or more). None of the tubes to which the 1 to 1,000 or stronger solutions were added, (some of which I have here for your examination), show any surface growth and if any growth has occurred along the needle track, it must have been soon arrested as even after several days it is impossible to say that any more bacteria are to be seen along the line of inoculation than were introduced by the needle in inoculating the tubes; and subcultures made therefrom remain sterile. Of the two treated with the 1 to 2,000 solution, one developed a mould fungus as in the case of the plates. Some of those treated with the weaker solutions grew, but less vigorously than the controls.

Another series of experiments was conducted with smear cultures the formalin not being introduced in this case until the cultures had been allowed to develop for from 24 to 48 hours. Sub-cultures made from these, after allowing the stronger solutions to act upon them for a few moments, remained sterile, but the weaker solutions were not so effective as when introduced at the moment of inoculation, a minutes exposure to the 1 to 1,000 solution not always being sufficient to sterilize the deeper layers of the surface culture.

Two or three drops of pure formalin applied to the cotton plug so that only the vapor came in contact with the culture was also found to be sufficient to sterilize both

fresh and 48-hour cultures of both diphtheria and anthrax as tested by subcultures.

Besides its use as here stated, I have used formalin to sterilize instruments and hands, to harden tissue and to render gelatine insoluble.

Gelatine, as is well known, melts at a comparatively low temperature, but after 24 hours exposure to formalin vapor it not only becomes insoluble in water, but loses its property of becoming liquid at elevated temperatures. Indeed I found by experiment that Schering's claim that it would not become liquid even when placed in the flame of the bunsen burner is true, the gelatine under these circumstances chars and may be burned but will not melt.

In bacteriology formalin is thus very valuable for class work for as pointed out by Hauser, quoted by Schering, plates, stab and smear cultures can be prepared and when the proper stage of development has been reached the vapor disengaged at the room temperature from a few drops of formalin will arrest further growth and render the cultures harmless in case of breakage without in any way altering their appearance. Colonies can also be permanently mounted by putting them with a little of the surrounding gelatine or agar on a slide, running a little of the melted medium under the cover, exposing this for 24 hours to formalin vapor and then spinning around a ring of cement to prevent evaporation ; or, what is better, as it prevents pressure from the cover, mounting in a deep cell after sterilizing with formalin.

For sterilizing instruments I have used a 10 per cent solution in alcohol. This so far as I can see has no bad effect upon the metal or cutting edge. For sterilizing my hands I have used a ½ per cent to 1 per cent solution. It is stated that formalin tans tissue and that when painted in full strength over the skin, the tissue involved separates without suppuration after a few days, leaving a rather deep eschar. I have accidentally spilled the pure

formalin upon my hands two or three times, immediately washing it off, without any injury to the skin but after the prolonged (½ hour) handling of tissues hardened in a 2½ per cent solution of formic aldehyde, one day I found the skin considerably roughened and somewhat benumbed for a day or two.

We have used the pure formalin in the place of nitric acid in a few cases of primary venereal sores and here some little pain was occasioned lasting longer than in the case of the acid, but causing no other bad results, the sores healing earlier considerably than in cases not so treated. Still on account of the pain I think it would be better to apply a diluted solution and repeat the application if necessary. The secondary symptoms in the cases of syphilis were of course not prevented; in those of chancroid there was no autoinoculation.

As regards the hardening and preservation of tissue I have used formalin to the exclusion of Mueller's fluid and alcohol recently and am much pleased with it. A block of tissue ½ or ¾ inch in thickness can be hardened in 24 hours or less in pure formalin without the great shrinkage in cells that occurs in the use of absolute alcohol. Even a 1 per cent solution acts quicker than Mueller's fluid and will preserve large specimens but as a hardening agent I prefer a stronger solution 5 per cent to 10 per cent which, except in the case of very loose tissue such as lung, renders a ½ inch block firm enough to cut within 48 hours. With a 10 per cent solution the blood and coloring matter seem to be fixed in the tissue so that comparatively little bleaching occurs; with a 1 per cent solution at the outset the bleaching appears to me to take place to nearly or quite the same extent as it does with alcohol.

I at first thought that the tissue rapidly fixed in formalin might be soaked in warm liquified gelatine or agar, imbeded in one of these media, exposed to the vapor of

formalin for a few hours and then cut but I failed to get thin sections in this way the imbedding material being too elastic to cut well. It seems necessary either to use a Valentine's knife or a freezing microtome for immediate examination or to replace the formalin by alcohol, and this by turpentine, cedar oil or chloroform and imbed in paraffine. Tissues so imbedded, cut well and the formalin does not affect their staining either in mass or upon the slide as regards any of the dyes that I have yet tried.

The manufacturer does not recommend the use of formalin internally, but states that the workmen exposed to its vapors in the laboratory have exhibited no evidences of its injurious effects. I have used a 1 to 500 spray upon my own throat without any bad effects and intend to use stronger solutions to test its action. The solution used had a peculiar penetrating sensation but not disagreeable. Inhalation of the vapor causes a slight stinging sensation somewhat similar to that of ammonia and is somewhat irritating but seems to produce no bad effects. The spray disengaged from pure formalin to disinfect some plates of diphtheria that I accidentally dropped on the floor and scattered over the laboratory while conducting my experiments with formalin irritated my eyes somewhat during the exposure, but the irritation ceased immediately after completing the disinfection leaving no unpleasant consequences.

As to the dosage in diphtheria I should expect good results from the application to the patches of a 1 or 2 per cent solution but would not hesitate to tentatively apply a stronger solution if necessary to check development of the bacilli. At the same time I should spray the throat with a 1 to 500 solution. Clinical experience, however, is necessary before formulating very definite conclusions on this point.

It is my opinion that formalin is destined to become an

important agent in histological and bacteriological technique; and its remarkable powers of penetration, its effectiveness and ease of application in solution, spray and vapor and its apparent harmlessness to tissues and fabrics in the quantities necessary to destroy bacteria, all combine to make it a valuable germicide.

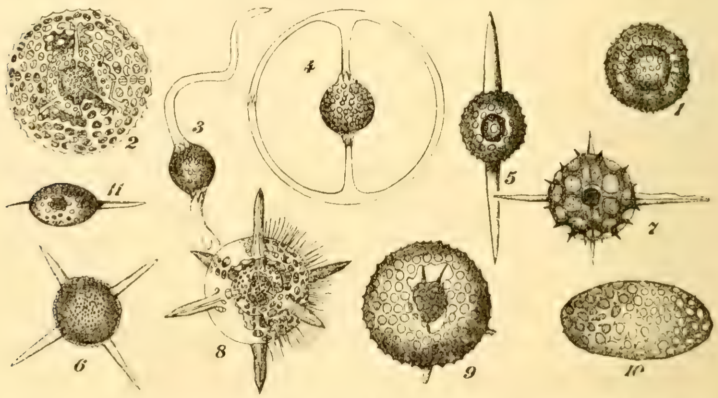
Radiolaria Classification Continued.

BY REV. FRED'K B. CARTER,

MONTCLAIR, N. J.

Continued from 1893, page 307.

In the *Stephoidea* there were 11 genera; in the *Spyroidea* 25 genera; in the *Botryodea* 5 genera; now we come to a sub-order in which there are 79 genera or almost twice as many as in all the other three sub-orders put together. This sub-order alone of the Legion NASELLARIA, or MONOPYLÆA more than equals the whole



of the legion SPUMELLARIA, or PERIPYLÆA in the number of its genera. Fortunately, however, it has four well marked sections, characterized by the number of the joints, and the distinction, MOUTH OPEN, or MOUTH CLOSED, which runs all through the sections, is also a great help in the determination of these forms.

VII. *Cephalis simple.*

SECTION A. CEPHALIS ONLY.

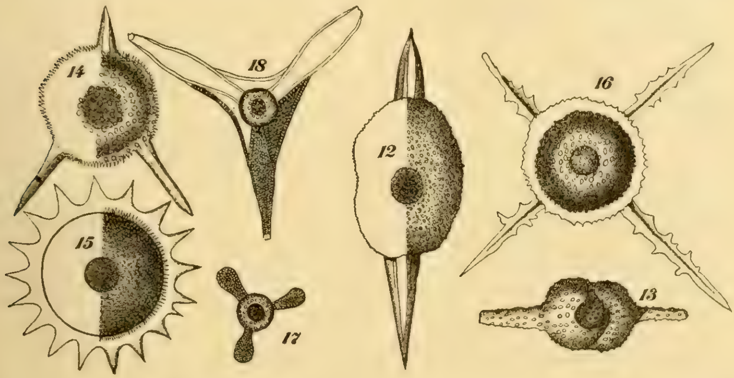
27.—Family: TRIPOCALPIDA. Three radial apophyses, i. e., ribs, wings, spines, or feet.

Three feet.	Horn. Three lateral ribs.	*Tripocalpis.*
Three feet.	Horn. No lateral ribs.	*Tripilidium.*

28.—Family: PHÆNOCALPIDA. Numerous radial apophyses.

BASAL MOUTH OPEN.

Radial ribs.	*Cinclopyramis.*
No radial ribs. Horn.	*Halicalyptra.*



No radial ribs. No horn.	*Carpocanistrum.*

BASAL MOUTH CLOSED.

Free columella in axis of shell cavity.	*Phænocalpis.*

29.—Family: CYRTOCALPIDA. No radial apophyses.

BASAL MOUTH OPEN.

Shell conical, dilated.	*Cornutella.*
Shell ovate, constricted. Horn.	*Archicorys.*
Shell ovate, constricted. No horn.	*Cyrtocalpis.*

BASAL MOUTH CLOSED.

Horn.	*Halicapsa.*

SECTION B. CEPHALIS AND THORAX, 2 JOINTED SHELL.

30.—Family: TRIPOCYRTIDA. Three radial apophyses.

MOUTH OF THORAX OPEN.

Three radial ribs enclosed in wall of thorax. No cephalic wings.

THREE THORACIC RIBS PROLONGED INTO 3 TERMINAL FEET.

Cephalis with horn. *Dictyophimus.*

THREE THORACIC RIBS PROLONGED INTO 3 LATERAL WINGS.

Thorax latticed. Horn. *Lithomelissa.*

Thorax latticed. No horn. *Psilomelissa.*

Thorax with spongy frame work. *Spongomelissa.*

Three radial beams free not enclosed in thorax.

Beams outside thorax. *Eucecryphalus.*

Three radial terminal feet on peristome. No ribs.

Feet solid. *Lychnocanium.*

MOUTH OF THORAX CLOSED.

Three divergent ribs enclosed in thorax.

Ribs in wall of thorax. *Sethopera.*

Three divergent free lateral wings on sides of thorax.

Three solid lateral spines. Horn. *Micromelissa.*

Three solid lateral spines. No horn. *Peromelissa.*

Three terminal feet (on base of thorax).

Three feet solid. *Tetrahedrina.*

Three feet latticed. *Sethochytris.*

31.—Family: ANTHOCYRTIDA. Numerous radial apophyses.

Radial ribs smooth, (rarely thorny), enclosed in wall of thorax.

(Cephalis commonly small, without horns).

Shell ovate. Mouth constricted. *Sethamphora.*

Shell pyramidal. Meshes simple. *Sethopyramis.*

Shell pyramidal. Meshes fenestrated. *Plectopyramis.*

Radial ribs *thorny,* (rarely smooth) prolonged into free *terminal feet.*

(Cephalis commonly large, with one or more horns). *Acanthocorys.*

No ribs in thorax. Peristome with free terminal feet. (Cephalis well developed with horn).

Six feet *Anthocyrtoma.*
Six feet *Anthocyrtis.*
Twelve feet, or more *Anthocyrtium.*
Feet outside constricted peristome *Anthocyrtidium.*
(Cephalis hidden. No horn). *Carpocanium.*

32.—Family: SETHOCYRTIDA. No radial apophyses.

MOUTH OF THORAX OPEN.

Thorax conical or campanulate. *Sethoconus.*

Thorax cylindrical or ovate, with truncate, constricted or tubular mouth.

One horn. Mouth simple. *Sethocyrtis.*
One horn. Mouth tubular. *Sethocorys.*
Two horns or a bunch. *Lophophæna.*
No horn. *Dictyocephalus.*

MOUTH OF THORAX CLOSED.

Horn. *Sethocapsa.*
No horn. *Dicolocapsa.*

SECTION C. CEPHALIS THORAX AND ABDOMEN. (3 JOINTED SHELL).

33.—Family: PODOCYRTIDA. Three radial apophyses.

MOUTH OPEN.

Three free limbs or wings on thorax (partly on abdomen).

No free external apophyses on abdomen.

Three wings of thorax solid. *Pterocorys.*

Free apophyses on abdomen.

Three ribs prolonged into 3 feet. Ribs and feet solid. *Theopodium.*

Three ribs prolonged into 3 feet. Ribs and feet latticed. *Pterocanium.*

Three free wings on thorax. Numerous feet. *Pterocodon.*

Free apophyses only on abdomen.

Three feet solid, simple. *Podocyrtis.*

Three feet solid, ramified. *Thyrsocyrtis.*

Three feet latticed. *Dictyopodium.*

MOUTH CLOSED.

Three lateral wings only on thorax. *Lithornithium.*

Three lateral wings prolonged into abdomen. *Theopera.*

Three wings only on abdomen.

Shell spindle-shaped. *Rhopalocanium.*

Shell 3-sided, pyramidal. 3 terminal feet. *Lithochytris.*

34—Family: PHORMOCYRTIDA. Numerous radial apophyses.

RADIAL RIBS IN WALL OF SHELL.

Ribs in thorax and abdomen. *Phormocyrtis.*

Ribs in abdomen only.

Peristome with free terminal feet. *Alacorys.*

Peristome smooth, without free feet. *Cycladophora.*

NO RADIAL RIBS IN SHELL WALL.

Abdomen cylindrical or ovate, not dilated. *Calocyclas.*

Abdomen truncate, conical or discoidal, dilated. *Clathrocylas.*

35.—Family: THEOCYRTIDA. No radial apophyses.

MOUTH OPEN.

Abdomen gradually dilated. Mouth wide.

Abdomen discoidal. *Theocalyptra.*

Abdomen conical, one horn. *Theoconus.*

Abdomen conical, 2 or more horns. *Lophoconus.*

Abdomen cylindrical. Mouth truncate.

One horn. Thorax and abdomen of equal breadth. *Theocyrtis.*

One horn. Thorax much broader, abdomen tubular. *Theosyringium.*

Two horns or a bunch. *Lophocyrtis.*
No horn. *Tricolocampe.*

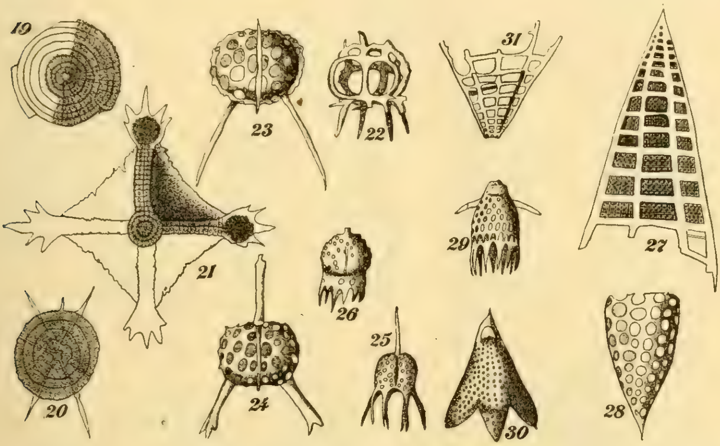
Abdomen ovate or inversely conical.

Mouth more or less constricted.

One horn. *Theocorys.*
Two or a bunch. *Lophocorys.*
No horn. *Theocampe.*

MOUTH CLOSED.

Horn. *Theocapsa.*
No horn. *Tricolocapsa.*



SECTION D. FOUR TO SEVEN OR MORE JOINTS IN SHELL.

36.—Family: PODOCAMPIDA. Three radial apophyses.

MOUTH OPEN.

Wings solid. *Stichopilium.*
Wings latticed. *Pteropilium.*

MOUTH CLOSED.

Three solid ribs. Cephalis with apical horn. *Artopera.*

37.—Family: PHORMOCAMPIDA. Numerous radial apophyses.

Shell ovate or spindle-shaped. Ribs prolonged into terminal feet. *Artophormis.*

38.—Family: LITHOCAMPIDA. No radial apophyses.

MOUTH OPEN.

Transverse strictures not connected by spiral line.

Shell conical or cylindrical. Mouth not constricted.

Conical. Horn. *Lithostrobus.*
Conical. No horn. *Dictyomitra.*
Cylindrical. Horn. *Artostrobus.*
Cylindrical. No horn. *Lithomitra.*

Shell ovate or spindle-shaped.

Mouth of last joint constricted.

Cephalis with horn. Last joint not tubular. *Eucyrtidium.*
Cephalis with horn. Last joint a long tube. *Eusyringium.*
Cephalis without horn. Cephalis with tube. *Siphocampe.*
Cephalis without horn. Cephalis without tube. *Lithocampe.*

MOUTH CLOSED.

Last joint rounded, without vertical basal spine.

Cephalis without horn. *Stichocapsa.*

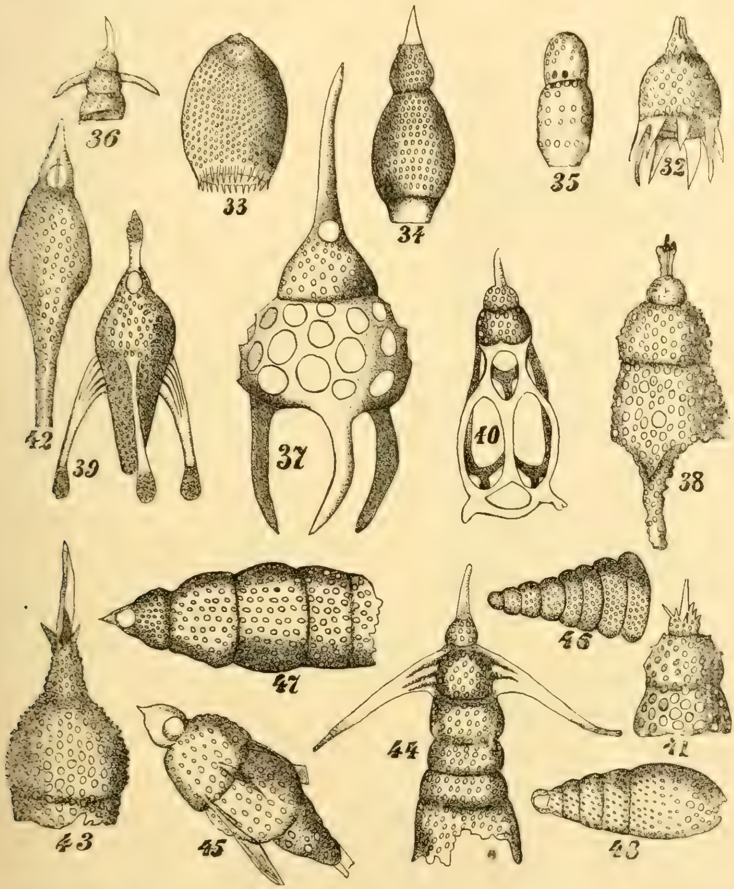
Last joint conical, pointed, with vertical basal spine.

Cephalis with horn. *Artocapsa.*

In addition to the 71 genera of the SPUMELLARIA and the 120 genera of the NASELLARIA, of which I have now given the classification, Haeckel mentions 3 genera of the Legion PHÆODARIA as being found in Barbadoes, namely *Mesocena*, *Dictyocha*, and *Distephanus* which belong to the family *Cannorhaphida;* but unless I am very much mistaken they have lately been ruled out as not being true Radiolaria. I have not thought it worth while therefore to do more than append the names.

Since writing the above I find it stated in an appendix to Haeckel's work that three more genera have been found in Barbadoes, namely, *Cortina*, *Stephanium*, and *Stephaniscus*; they belong to the *Stephoidea*, the first and second to the Family *Stephanida*, the third to the Family *Se-*

mantida. The presence of basal feet distinguishes *Cortina* and *Stephanium* from *Dendrocircus* and *Zygocircus* and the number of feet separates them from each other, *Cortina* having three and *Stephanium* four. *Stephaniscus,* of the Family *Semantida,* is distinguished in the



same way from *Cortiniscus,* the latter having but three feet while *Stephaniscus* has four. This increases the total number of genera in Barbadoes to 194.

ERRATA.

In the Journal for November, 1893, pp. 306-7:

Family 21, D. For *Tanrospyris* read *Taurospyris.*
Family 22. For *Pharmospyrida* read *Phormospyrida.*
Family 26. For *Rylobotryida* read *Pylobotryida.*

LIST OF ILLUSTRATIONS.

SPUMELLARIA.

1. Carposphæra entactinia.
2. Thecosphæra inermis.
3. Xiphostylus anhinga.
4. Saturnalis cyclus.
5. Stylosphæra liostylus.
6. Staurosphæra simonis.
7. Staurolonchidium perspicuum.
8. Hexacontium asteracanthion.
9. Haliomma oculatum.
10. Cenellipsis ehrenbergii.
11. Druppatractus lævis.
12. Spongatractus pachystylus.
13. Cannartidium amphicanna.
14. Triactiscus tripodiscus.
15. Heliodiscus humboldtii.
16. Staurocyclia serrata.
17. Trigonactura rhopalastrella.
18. Hymenactura trigona.
19. Porodiscus concentricus.
20. Stylodictya gracilis.
21. Histiastrum coronatum.

NASSELLARIA.

22. Tympaniscus fibula.
23. Dipospyris mystax.
24. Dendrospyris stylophora.
25. Petalospyris foveolata.
26. Patagospyris confluens.
27. Cinclopyramis cribellum.
28. Cornutella mitra.
29. Eucecryphalus campanella.
30. Sethochytris barbadensis.
31. Sethopyramis quadratella.
32. Authocyrtis mespilus.
33. Carpocanium coronatum.
34. Sethocorys armadillo.
35. Dictyocephalus crassiceps.
36. Pterocorys apis.

37. Podocyrtis triacantha.
38. Dictyopodium eurylophos.
39. Rhopalocanium ornatum.
40. Cycladophora hexapleura.
41. Lophoconus apiculatus.
42. Theosyringium tubulus.
43. Lophocorys acanthocephala.
44. Stichopilium macropterum.
45. Artopera loxia.
46. Dictyomitra articulata.
47. Eucyrtidium montiparum.
48. Lithocampe clava.

NOTE.—The authority for the above names is as follows: Nos. 5, 9, 23, 25, 28, 32, 33, 37, 38, 39, 47, 48 were named by Ehrenberg; all the others were by Haeckel.

LETTERS TO THE EDITOR.

NOTE.—*This column is open to all correspondents who write upon the topics enumerated under "Problems," or who give other information of interest. The fact that a problem has been answered once need not deter our friends from making additional comments. To facilitate reference, correspondents should cite the number as well as the page on which have appeared letters and queries to which reference is made. The editor is not responsible for the views of others published in this periodical.*

Blood Corpuscles.—Relative to my article on the measurement of blood corpuscles which you have done me the honor to republish in your January number, 1894, I desire to say that paper does not reflect my matured judgment upon the question involved My latest views upon the subject formed upon more extended observation, and experience will be found in the September number of the Medico-Legal Journal, 1872, published at 57 Broadway, New York. M. D. EWELL, M. D.

Bird Parasites.—I am especially interested in the parasites of birds and should like very much to see the key to species proposed to be published by Prof. Osborn. I use the large-size Beck National Monocular and I am a'so interested in photomicrography. If I get into the parasites of birds I will send you what I desire to print. R. W. SHUFELDT, U. S. A.

TACOMA, D. C., *March* 29, 1894.

The Journal grows in interest and value with each number.
W. LIGHTON.

OMAHA, NEBRASKA, *March* 15, 1894.

EDITORIAL.

Prof. Gage's New Book on the Microscope.—In the March number of the JOURNAL a brief notice was given of the fifth edition of "The Microscope and Microscopical Methods," by Prof. S. H. Gage which had just appeared. A fuller and more careful examination of this book has shown it to be possessed of such a high degree of excellence that we wish to call the attention of our readers to the fact that America has now produced a work on the microscope which will compare more than favorably with any of the productions of European writers on this subject. Although the book was prepared primarily for the laboratory student, it contains such a vast amount of information concerning every part of the microscope and its functions that it deserves a place among the reference books of science. To the student or physician who wishes to study the microscope we can recommend it with a feeling of assurance, that if it is carefully read and its directions followed no difficulty will be experienced in the manipulation of this instrument. It is a practical book. It is concise and especially clear in its definitions of the various technical terms necessary for a working knowledge of the microscope. We are more than delighted that such an admirable work has come within the reach of everyone, and we hope that the diligence and enthusiasm which has characterized the author in his most successful labors to render more intelligible the "mysteries" of the microscope, may be imparted with equal success to all of its readers. With such a practical adjunct as this volume in which is brought together all that is essential from the scattered literature on the subject the student of the microscope must succeed if persistent in his labors. The JOURNAL is much gratified, in its ever persistent efforts to present the needs of a knowledge of the microscope, and its practical application to the attention of students, teachers and physicians to be able to recommend this masterly production of Prof. Gage. In fact, every microscopist should possess a copy of this work and every student of the microscope should read it carefully. We would emphasize in reference to all subjects in which the microscope is used, the comment of the Rev. W. H. Dallinger on the former edition of this work. "In short, this treatise lays the foundation for a

thorough microscopical training entirely adapted to the wants of medical students." We are glad to announce that, notwithstanding the considerable size of the book and its many illustrations that it is placed upon the market at a price within the reach of all.

The Earthworms.—Those interested in the study of Oligochæts will find in the Memoirs of the California Academy Science, II, No. 3, just issued, the descriptions of several new species and a series of 12 large plates which contain most interesting anatomical figures. The paper constitutes a monograph on the family Eudrilidæ.

MICROSCOPICAL APPARATUS.

Section cut Versus Natural Preparations.—Staining and section cutting is one of the most valuable methods in use in microscopical investigation, but it should be avoided in many instances where the object can be examined in a satisfactory manner without undergoing this process.

The hardening process in alcohol of necessity contracts the soft tissues, while the knife divides some objects in a manner to render them hardly recognizable, in fact, in such cases the specimen is transformed from its true and natural aspect, to something false and unnatural, and it is a question, if due allowance is always given, in the examination of hardened section preparations to these facts.

This is very apparent in two mounted preparations now before me, one a preparation of Trichinæ in the muscle of the hog, hardened, stained and cut in sections; the second is the same object, simply pressed, dried and mounted, as I will presently describe.

A glance is sufficient to detect, in this case the value of the second method over the first. The former shows the trichinæ much decreased in size, of an unnatural color, and their forms mangled and distorted, while in the latter case they have all the appearance of nature, as seen in a fresh preparation—the animals and the cysts are perfect and lie in their usual place.

The latter preparations are made by taking very fine particles of the fresh muscle, using for the purpose a sharp pair of scissors, and placing them between two pieces of thick glass, and pressed

with all the strength possible, giving a slight lateral movement at the same time. If then held up to the light, the meat will be found almost transparent with a slight opalesence. If now placed in the compressor frame, and screwed down, the preparation will be ready for microscopical examination. In about 24 hours or more, the water in the specimen will evaporate, and if the glasses be removed from the compressor and separated, the preparation will be found to be a very thin film, transparent and dry, which can be removed and cut in suitable pieces, and mounted in the usual way, in its natural condition or stained. The compressors mentioned are those used by the meat inspectors of the U. S. Bureau of Animal Industry and made by Bausch & Lomb. They are illustrated at No. 1662 of their last catalogue.

The glasses are 3x2 inches, and about 1-10 of an inch thick, and will bear a considerable pressures without breaking. They offer great facilities to microscopists, as there are a large number of objects that require pressure during preparation, especially in Botany and Insect life, and from the size, quite large objects can be introduced. It is cheap costing only two dollars including the two glasses. JOHN MICHELS.

The Pelham, Boston, Mass.

MICROSCOPICAL MANIPULATION.

Microphotographs by Lamp-light.—Dr. Borden, U. S. A., of Fort Adams, Newport, has taken some fine microphotos of bacteria with lamp-light. He uses a Laverene three inch magic lantern with four inch condensors. The lantern objective is removed and the middle wick only lighted. The substage condensor is focussed accurately on the front surface of the stereopticon condensor. This gives light with a ½ inch oil immersion so that a magnification of 1000 diameters can be focussed on the screen. With a 1-5 inch objective he uses a one inch objective on the substage. He uses a Cramer's Isthochromatic rapid plate and develops with hydrochromion. This plate is specially prepared for lamp work and gives the best results of many different makes. A 1½ to 2 minutes exposure is needed for 1000 diameters.

Reflected Light.—It is well known that light reflected from white clouds is one of the very best for illuminating the microscope. Mr. Geo. Rust writes us that when white clouds are wanting, he moistens the finger with watch-oil and spreads an even but very thin coat over the face of the mirror which produces much the same results.

MEDICAL MICROSCOPY.

Too Busy to Use a Microscope.—A late lecturer at a medical college on "medical science as now enlightened by microscopic research" was told by a confrere: "We are so busy that we have no time to go into such things."

In other words, we practice in the dark and without the best results, simply because our time is fully occupied. We make our profession profitable for ourselves if not for our patients.

To be sure, they think they have the services of those fully up to, if not above the average physician. Some patients think we are the best of our cult. At any rate the physical signs of business comply with the ethical standards of professional success, but we have not time for the microscope. If once in a year or two, we want urine examined, we send specimens to the apothecary or some medical expert who never sees the case, making a laboratory and not a clinical investigation.

So it is that your expectations are not to be realized. We are too busy to come up to this standard.

My friends look out! Now that the use of the microscope in disease is taught to first year students in such colleges as Mass. Institute of Techology under the head of biology, the time is coming that such graduates will mould public opinion so that you will have to use the microscope clinically. Sick people may get an idea that the physician is to cure if possible, your patients may prefer cure than death because you are too busy to properly study their cases. The sick are entitled to the benefits of all means of clinical knowledge and those who keep their patients from these benefits are, to put it mildly, not honoring their profession as they should.

We do not claim too much for microscopy but it is not right to wholly ignore the grand clinical work the microscope is now doing for the sick. Many are alive to-day who but for the clin-

ical teaching of the microscope would have died years ago.

Lung fibers in phthisis sputum discovered before 1826.—This is not an Amercan teaching. In 1826 Prof. Schroeder Van der Kolk of Utrecht, Holland, published his "Observationes, anatomici, pathologici et practici argumenti." These observations include lung fibres in the sputum of phthisis. He died May 2, 1862 The authority for this is an autograph letter dated Feb. 24, 1894 by Dr. G. P. Tienhoven of the Hague, physician to the Queen of the Netherlands. Honor to the Dutch! How much we owe to them not only for the microscope but also for the accurate mode of detecting the breaking down of the lungs not only in tuberculosis but also in fibroid phthisis and fatty degeneration.—E. Cutter, M. D.

BIOLOGICAL NOTES.

Irritability of Phycomyces nitens.—Dr. Elfving has published a paper concerning the effect of different bodies upon the sporangia-bearers of *Phycomyces nitens* (fur Kenntniss der pflanzlichen Irritabilitat. Sep. from Oefversigt af Finska Vel.—Soc. Foerh. Haeft XXXVI, 1893). This action is either attractive or repulsive; from a distance of a couple of cms, iron and zinc will cause the sporangia-bearers to curve so that the curved side faces the active body.

Errera explained the movements of said organs as hydrotropism. Elfving publishes new experiments: If iron acts as a hygroscopic body, we may expect to see the phenomenon most plainly when the fungus is put under the influence of highly hygroscopic bodies like calcium cloride and Ka.—But the sporangia-bearers were not attracted by these bodies.

A very hygroscopic plate of gypsum (80x35x10 mm.) dried by 100° C., and placed in an atmosphere saturated with water, in the neighborhood of the sporangia-bearers had no effect whatever upon the latter and condensed 1,665 gr. of water, while an iron plate (surface 4950 mm^2) produced the effect mentioned above and condensed only 3.5 mgr. of water.

Elfving assumes that these phenomena are caused by molecular movements. Highly polished steel and platinum have very little effect upon the sporangia-bearers, but if these bodies, for a long time, are exposed to direct sunlight, they become active, i. e., they are brought into such a condition that they at-

tract the sporangia-bearers. This condition only lasts for some hours, and then it disappears.

We know that a number of nonphosphorescent bodies emit rays of light after having been under influence of light. The duration of this condition varies from a few minutes to 24 hours. Metals such as steel or iron are not phosphorescent, but we have here a new form of phosphorescence, which might be called dark phosphorescence. It is the light and not the heat which produces the said effect upon the metals; the color of the rays do not seem to have any influence upon producing the effect described above, in the metals.

Zinc becomes active by heating alone, and when experimenting with this body, Elfving found that it becomes positive thermotropic. On copper, cobalt, nickel, tin, lead and glass heating like above alone did not produce the activity, these metals being heated until they are nearly melting, and then allowed to cool so far that the hand could not feel the heat.

Elfvings conclusion is: Es scheint mir dann wenig befremdend anzunehmen dass auch Molekularschwingungen, welcke den Rorpern selbst innevohnen oder irgend eine in denselben slattfindende Veranderung begleiten, ahnliche physiologische Wirkungen hervarrufen Konnen. Was speciell die Metalle betrifft, zeigt uns ja auch die Metallotherapie Wirkungen, die entschieden fur solche sprechen.

MRS. DORA BAY.

MICROSCOPICAL NOTES.

T. D. A. Cockerell is now curator of the museum at the Island of Jamaica.

G. W. Rafter, one of our valued contributors, has recently written a book on Sewage Disposal which has been published by Van Nostrand. We have not yet seen a copy.

MICROSCOPICAL SOCIETIES.

Washington, D. C., L. M. Mooers, Secretary.

April 10, 1894.—In spite of a heavy rain storm, seven members were present, Dr. Gibbs in the chair. The Soiree Committee reported the arrangements for a Soiree at the High School building May 8th. Mr. Smiley showed photographs of blood

taken by Dr. E. Cutter with the celebrated 1-75 inch objective. For comparison, Dr. Reyburn showed a negative of blood taken by the late Dr. Woodward with Zeiss 1-12 inch objective.

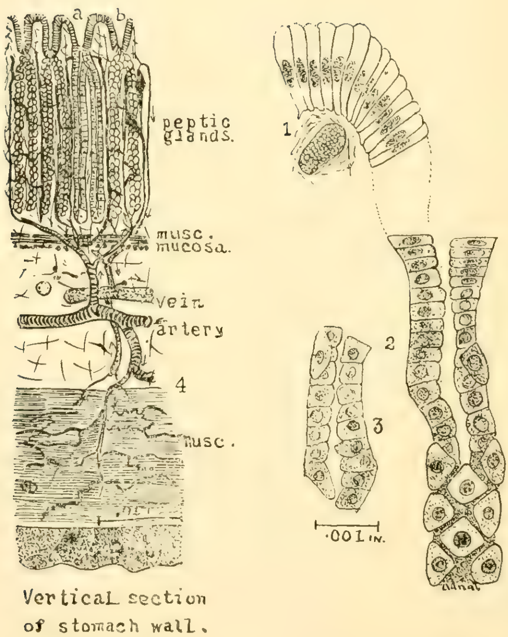
Dr. V. A. Moore presented a paper upon Distinguishing the Typhoid bacillus from the Bacillus coli. They cannot be differentiated morphologically even with the aid of the microscope. He has made extensive observations upon the flagellæ in the hope to do so but has failed. It is found however, that when introduced into a 1 per cent solution of glucose peptonized boullion the B. coli causes fermentation with the formatiou of gas and the typhus does not. Fermentation tubes were exhibited proving this fact. By request, Dr. Moore also described the method of plate culture for isolating different species of bacteria and of securing pure cultures for use in experimentation. He exhibited the Bacillus of typhoid fever under the microscope.

Dr. Alleger explained his method of testing suspected water for typhoid germs. If water contained, as it might, a score of other germs in connection with the typhoid, the plate-culture method of separating them would require long and tedious operations. But it has been found that at a certain degree of temperature many of these will remain inert, while with an acid medium and temperature36°C. the typhus and only a very few others will grow. By producing these conditions a great number of harmless species are eliminated from the work and the method of finding the deadly typhus much shortened.

NEW PUBLICATIONS.

Investigations on Microscopic Foams and on Protoplasm. By O. Butschi. 8° London, 1894. Price 18s.

This is a translation by E. A. Minchin of Merton College, Oxford. It covers experiments and observations directed towards a solution of the question of the physical conditions of the phenomena of life. Prof. Butschli discovered all gradations between scattered vacuoles and a completely alveolar or reticular structure. He then succeeded in making foams which under the microscope presented the reticulated appearance of protoplasm. He also discovered streaming movement and actual progression in these foams. Properties heretofore attributed only to living protoplasm are thus found in (living?) foam.



STOMACH WALL OF A CAT AND DETAILED VIEW OF THE EPITHELIUM IN ONE OF THE TUBULAR GLANDS. DRAWN WITH CAMERA LUCIDA.

FOR EXPLANATION SEE PAGE 146.

THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. MAY, 1894. NO. 5.

A Method for Orienting Small Objects for the Microtome.*

BY W. McM. WOODWORTH,

CAMBRIDGE, MASS.

In studying the embryology of *Polychœrus caudata*, an acœlous Turbellarian, I experienced great difficulty in obtaining sections in definite planes, owing to the extreme smallness and nearly spherical form of the early stages. The difficulty lay in controlling the plane of section in relation to the axes of the object. The embryos measure only about 0.224 mm. in diameter, and all orientation must be done under low powers of the microscope. I tried orienting in paraffin, which was kept fluid by means of hot water circulating through a combination Stricker's gas and warm stage into the central well of which iced water could be quickly introduced. I hoped thus by the sudden cooling of the paraffin to fix the object in the position into which it had been brought by means of needles. This method proved useless, for owing to their lightness and spherical shape the objects could not be kept long enough in one position, being moved about by the convection currents in the hot paraffin. The method suggested by Born (Zeitschr. f. wiss. Mikr., Bd. V. p. 436, 1888) was also tried, but was not applicable to objects so small and round, it being impossible, in early stages, to determine the position of the poles of the egg owing to the fact that the grooves of

*From Bull. Comp. Zool., Harvard College, XXV, 3.
To orient: To define the position of.—Webster.

the cleavage planes become filled with paraffin and are thus obscured. The method is useful, however, when the embryos have developed a characteristic shape or symmetry that is visible through the coating of paraffin. I was still at sea with my young stages when Dr. William Patten told me of a method employed by him, in experimenting with which I developed the method which is the subject of this communication.

The method depends upon the use of paper having a surface of raised parallel lines, or in other words, having a grained or rep-surface. Writing paper of this kind is made in various sorts and can be had in the market. The best kind is that which is known as "linen cloth," and is made in imitation of some coarse fabric. It bears series of parallel raised lines intersecting one another at right angles in imitation of the woof and warp of some woven fabric. The surface of the paper is thus divided into minute squares, the meshes representing the spaces between the imitated threads of the fabric. If such paper is not to be had, any paper that has a distinct grain will answer, provided the lines of the grain are straight and parallel.

Cut a rectangular strip of the paper so that the cut edges are parallel and perpendicular to the direction of the grain (I use strips about 5x15mm.) and paste it smooth to a glass slide by means of a solution of gum-arabic. The rougher side of the paper, i. e. the side showing the grain more distinctly, should lie uppermost, or exposed. When the gum has dried, the exposed surface of the paper is coated with a thin film of the gum-arabic solution, which is best applied with a brush. When this is quite dry, the gummed surface of the paper is coated with a thin layer of collodion. This should be ordinary flexible collodion diluted with three parts of ether, and should be applied with a small brush so as to produce a very thin film. The coating of collodion

should not be added until immediately before use, for, if allowed to remain too long, it is liable to crack and fall away. The slip of glass thus prepared with the strip of paper covered with its two films of gum and collodion is then ready for use.

The object to be oriented, which has been previously cleared in turpentine, is now drained of the superfluous oil by means of bibulous paper, and brought on the point of a needle to the surface of the paper prepared as above, and adjusted under the microscope so that the axes of the object have any desired relation to the lines on the paper. If care be taken to drain off all of the turpentine from the object, it will stick to the surface of the collodion in any position in which it is placed, so that an oblong or ovoid object can be placed either on end, or so that its chief axis will make almost any angle with the plane of the paper. When the object is suitably oriented, the whole slide is exposed under a bell glass for a few seconds to the vapor of ether. This softens the collodion, which upon drying holds the object fast in the desired position. The object is then covered with a drop of turpentine, and the slip of glass with the attached paper is placed in the paraffin bath.

To imbed the object in paraffin, the slip of glass bearing the object is removed from the bath, and a mould is built upon it about the paper in the ordinary way, by means of bars of metal arranged in the form of a rectangle, and filled with fluid paraffin. The mould should be as nearly as possible of the same size as the strip of paper, that is to say, the sides of the mould should coincide with the edges of the paper. When the paraffin has cooled, the metal guards are carefully removed, and the paraffin is cut away until the edges of the paper are exposed. This is an important preparation for the next step. The glass slip with the attached block of paraffin is then put into a vessel of water. The water working

into the paper from its edges dissolves the gum-arabic, and the block is thus detached from the glass. The film of gum between the collodion and the paper is also dissolved, so that the paper can then be removed, leaving the block of paraffin with the imbedded object. The object thus lies close to the lower surface of the block, over which is the delicate film of collodion and the imprint of the grain of the paper. One face of the paraffin block, which is a plane surface, thus bears a cast of the grain of the paper, the lines of which have definite relations to the axes of the imbedded object. It is now a simple matter to orient the block in the microtome so that the lines on the face of the block will have any position in relation to the plane of section. A whole series of objects can thus be oriented on one strip of paper; and with a fine pen numbers referring to notes and drawings can be marked on the collodion surface adjacent to each object. These marks will appear on the face of the paraffin block after imbedding. The film of collodion on the face of the block in no way interferes with making good "ribbons," for, owing to the fact that the collodion was much diluted, the film is so fine that it offers practically no resistance to the knife in cutting, and does not endanger the specimen.

Although necessitating a rather long description, the method is extremely simple. The chief advantages are: (1) the easy orientation of objects when filled with some clearing medium, by which details are made more visible; (2) the conspicuousness of the objects through all stages of the process; (3) the saving of time in being able to imbed many objects at one operation; and (4) the absence of any necessity for haste. The method has been tested by others with various objects, and always with success.

Dr. Hauser.—A chair of Bacteriology is to be established at Erlangen and Dr. Hauser will probably occupy it.

The American Microscopical Society.

By ARTHUR M. EDWARDS, M. D.,

NEWARK, N. J.

You ask me to give you what I know of the American Microscopical Society. This is all I remember or can know:

At my house, 49 Jane street, during the years 1864 and 1865, there used to meet a trio of enthusiastic microscopists consisting of C. Van Brunt, G. S. Allan and myself to consider natural history generally but the microscope particularly. At one of these meetings, it was proposed by Van Brunt I think, to get together the microscopists that lived within hailing distance to form a society. Van Brunt and Allan did not live at that time in New York, so it was proposed that I should call the meeting to be held at my house. This I did and put a notice in the paper that "a meeting will be held this evening at the house of Mr. A. M. Edwards, 49 Jane street, for the purpose of organizing a society for the advancement of microscopical research." I have not kept any date of this but it is a scrap from the paper of April 8, 1865. A goodly number of gentlemen were present who were interested in natural history more especially microscopy. I remember besides Van Brunt and Allan those present were R. K. Browne, S. Jackson, J. W. S. Arnold, S. A. Jones, W. H. Atkinson and several others. We organized the society under the name of the American Microscopical Society, for it was the first to be formed in America. We adjourned to meet at the same place one week from that date and a committee was appointed by the chairman, Dr. R. K. Browne, to draw up a constitution and by-laws. April 15, 1865 is a memorable day in the minds of all, for on Good Friday; April 14, 1865, President Lincoln had been assassinated at Ford's Theatre in Washington, and had died at the house opposite,

where he was taken from the theatre. A gloom overspread the whole land and we met together to mourn our loss and to organize a society. The committeemen who were appointed were S. Jackson, S. A. Jones and myself. I drew up the constitution and by-laws and proposed the seal which was a diagram of the refraction of light with the motto *Fiat lux*, the motto being the suggestion of Dr. S. A. Jones. We then organized and signed the constitution, I being the first to sign. Officers were then elected, being A. M. Edwards, President; R. K. Browne and Mr. G. S. Allan, Vice Presidents; C. Van Brunt, Treasurer; S. A. Jones, Secretary; S. Jackson, curator; and J. W. S. Arnold, Librarian. We met at the houses of several members until we got too large in numbers, when we met at a general public meeting room. This was at last the Mott Memorial Building, 64 Madison Avenue, New York. There I went out of the presidency and was succeeded by J. E. Gavit, President; R. Dinwiddie and W. H. Atkinson were Vice Presidents; A. M. Edwards, Corresponding Secretary; J. H. Hinton, Recording Secretary; T. D'Oremiealx, Treasurer, S. Jackson, curator; and J. W. S. Arnold, Librarian. The society went on swimmingly until one person was admitted. I do not know by whom, but as soon as he got in there was trouble, at least several of the members went out. I was one of the first to go. The society met and met, decreasing in interest until it again met at the members houses. It dwindled down in numbers and lost its interest until it ceased—when I do not know for I had ceased to be a member and had moved out of the city to reside in Newark, N. J. This is all I can remember or care to remember of the American Microscopical Society as it was then called. It was organized under the laws of the state of New York, December 22, 1866, under the name of the American Microscopical Society of the city of New York.

Studies of the Histology of Various Mammalian Tissues.—II.

By HENRY L. OSBORN,

ST. PAUL, MINN.

Continued from page 70.

THE ALIMENTARY CANAL.

In preparing the stomach or intestine for sections it is important that a clean surface of the inner lining be exposed to the action of the reagents used in preservation, this can be attained by carefully washing the inner surface of the organ with slightly warmed salt solution, made by dissolving six parts of common salt in one thousand of water. Pure distilled water should not be used as it causes rupture of the wall of the cells and thus destroys their natural shapes. After the surface is thus cleaned pieces should be hardened either by the alcohol method which is perhaps the most convenient or if preferred by the chromic acid, or other common mode. Injected specimens are necessary to give one a realizing sense of the structure, specimens injected with a gelatine mass can be hardened in the ordinary way for non-injected tissues and have the advantage of preserving and displaying both the vascular arrangements and the cell structure.

THE ALIMENTARY TUBE presents a roughly similar construction in all parts, but is different both structurally and physiologically in different places in its details. It is in general a double tube one inside the other, the outer tube is muscular the inner one is composed of gland-cells, between the two is a connective tissue coat in which the principal nerves and blood-vessels run. The three layers are known as the muscular, the submucous and the mucous coats named in their order from the outside inward. The mucous and the muscular are the active agents in the work of the organ while the middle coat is merely accessory. In addition to the organs

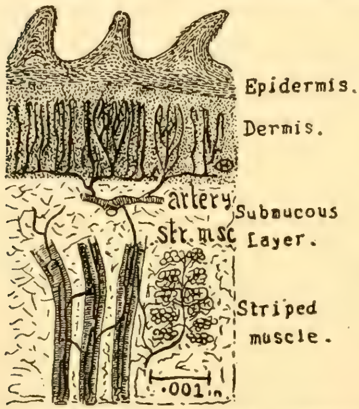
in the direct line of the tube there are others which are separate from it anatomically but directly connected with it by ducts or passages which bring the secretions of these connected organs into the tube itself. The salivary glands are organs of this sort as well as the liver and the pancreas. In the case of all of these organs two functions are notable namely motion and secretion, in the tongue in addition there is the function of sensation, both of taste and touch, in a high degree. The study of the structure of the organs has to be guided by the facts of their use and the two aspects of their study are thus complementary the one of the other.

THE TONGUE as a whole in the mammals is larger behind where it is attached to the floor of the mouth chamber and to the throat, and elongate in front. It is composed of muscular tissue which material is covered with epidermis, the character of which latter is peculiar on the upper surface, being thrown into certain "papillæ" or elevations of various shape in different parts. The muscular part of the tongue can be compared with the outer coat of the alimentary tube and the epidermis with the mucous coat, the two layers are not however loosely related as they are in the rest of the tube but are in the closest relation, the arteries lie in the level between the outer and the muscular layers which thus corresponds with the submucous coat. The extreme mobility of the tongue is not matched by the power of any other muscular organ in the body, and the range of motion is also enormous, this is attained through the presence in the bulk of the structure of sets of striped muscular tissue each one of which is composed of fibres running parallel. These fibres run in many different directions, some crossing from left to right, others running lengthwise and still others running vertically to the surface. The contractions of one or other of these sets would give rise to resultant motions of the organ. In

the cut two sets of fibres are shown, one the vertical are cut lengthwise in a vertical section and exhibit their length in the field of view. The others, transverse ones, are cut cross-wise and only the ends of the fibres are to be seen. In other parts of a section still other sets of fibres, those running lengthwise, would appear. The length of the fibre is not shown in the section and rarely if ever can be seen in sections, but the breadth is shown and can be measured by the scale shown in the figure, a fibre is thus seen to be less than one one-thousandth of an inch in diameter. The relation of the fibres to their vascular supply is also shown in the figure. The main artery is seen in the level next to the dermis, from it articles branch out in two directions, one set running up into the papillæ of the dermis, and the others to the muscle fibres, where their capillaries follow the length of the fibres so that all parts are in close relation to the supply of nutriment. The distribution of the blood to the sets of fibres or fibre bundles is shown in the cross-section of the bundle. If the section were to show all the parts of an organ it would include a view of a nerve fibre running to and terminating in one of the fibres. The tongue is one of the best objects on which to study the histology of striped muscle, for here the fibres are not so compactly arranged as they are in a muscle of the skeletal system.

The surface of the tongue is covered with a layer which is clearly divided into two parts, the upper is clear, nearly transparent in section and does not stain deeply if at all and this is also completely destitute of capillary blood vessels, and a layer below this which is the vascular layer and is composed of rounded cells with more decided affinity for carmine. The blood vessels in the deeper layer are derived from large trunks which are clearly visible in the stratum beneath the skin, and run up in the form of loops which are vertical to the horizontal supply vessels underneath. In some cases the capillaries are not

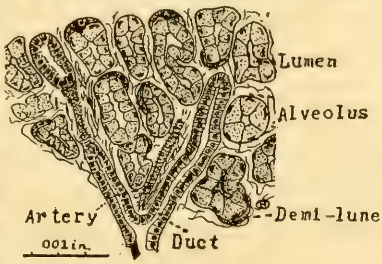
simple vertical vessels, but are in the form of an oval cluster. These vessels really underlie the cells of the epidermis which receive blood from the capillaries by diffusion from the spaces beneath them. This of course renders it easier for the lower layers of this structure to receive blood and the cells in the upper levels are at a disadvantage in this respect. The consequence is that they die, but not until after they have secreted a horny material allied to the chitine of the insect skeleton. This makes the cells extremely durable and thus enables them to endure the conditions that are placed on them by their exposed situation. Living delicate cells exposed on the tongue's surface to such changes as for instance from a dish of ice cream to a cup of boiling coffee would probably give up the struggle for life in complete despair. The outer surface of the tongue is covered with these dead cells, many layers deep, they are not in the form of a flat surface as in the œsophagus, but are, in the cat and in all the carnivora, in the form of sharp-pointed elevations, placed so as to point backward. These "filiform papillæ" are of use to the cats in the manner of a file rasping portions of the meat which constitutes their diet from the bones on which it grows. In the case of the lion these papillæ are so stout and strong that a single lick of the creature's tongue across the back of a persons hand would drag away with it all the skin down to the bare bones. In the tongue of the cat they can be also clearly seen. In the human tongue similar papillæ are present but they lack the sharply pointed shape of the carnivore being flattened down on top. In all cases



Vertical section of the tongue of a cat injected.

such papillæ are formed of epidermal cells flattened down and pressed together the shape of the mass being imparted by their formation on the surface of dermal papillæ of different kinds. In order to demonstrate the sensory function of the tongue other sections in special parts of the organ are necessary.

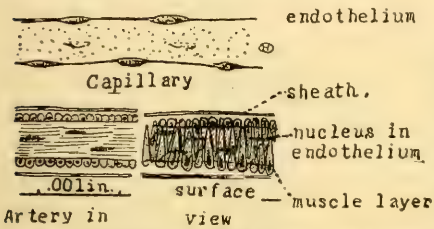
THE SALIVARY GLANDS are also a part of the mouth though not located in it. There are several different ones, the most convenient one to study is the "parotid" which as its name implies lies beside the ear. It lies a little below that organ. Sections properly hardened and sliced will show the parts displayed in the adjoining figure, the organ in the main is a mass of rounded areas of various shape, in places elongate passages are visible, and perhaps a connection between the rounded areas and these passages will be discernable. Besides these two there will also be seen in places, views of arteries. Some veins will perhaps also be discovered but unless the section is injected the vascular structure will be only very inadequately shown. The nerves will be undistinguishable from the areolar connective tissue, spread out everywhere between the divisions of the gland. Since the glands are of the racemose type, the terminations of the ductules will appear as closed portions, with a cavity the "lumen" in the centre. The section here represented is such a slice as would be obtained by a cut through the organ in any plane vertical to the surface. At first the section will appear to the untrained eye as a confused mass of faintly colored spots, but by degrees the eye will analyze the confusion and the principal structural elements in their relation will be discerned. The section here repro-



Parotid gland of cat.

duced is a camera-lucida drawing from an ordinary section of the organ. The bulk of the organ is composed of the alveoli or final parts of the system of ducts, one of the ducts with its branches is seen, cut for a part of its length, it is not seen to open into the alveoli, because a view of this is rarely attainable in sections, though it is sometimes to be seen. Running beside one of the branches of the duct is seen the artery. The spaces between these parts are occupied by the connective tissue or by the capillaries or nerves, which are hardly distinguishable from the areolar connective tissue. The cells of the alveolus are of two sorts. There are first the "serous cells" which compose the bulk of the organ. These cells are shaped like spherical wedges and hence appear triangular in sections. The cells do not wholly fill the alveolus, each one is a trifle shorter than half the diameter of the alveolus thus leaving a small space in the centre, called the "lumen." The lumen is in communication with the hollow passage in the centre of the duct. The serous cells are of a particular shape and character, they are comparatively clear, and their nucleus is small and flattened in the plane of the base of the cell, it also lies near to the base, so that in some cells it is hardly noticeable at all. There is another kind of cell in salivary glands called the "mucous" cell, this is not found in the parotid gland of the cat, it is characterized structurally by the fact that the nucleus is spherical and occupies the centre of the cell. The bases of some of the cells of the alveolus are clasped or covered by dark deeply stained cells, called "demi-lune" cells, there are a few only of these to each alveolus, and hence they are not seen in all the parts of the section. The duct is also lined with cells which become evident in sections after study, the side walls of the cells are rarely seen, but the nuclei are regular in position and from their shape and position one must infer the position of the side wall. These cells are tall and expose

a much slighter area to the fluids in the spaces of the surrounding tissues, than do the alveolar cells, they stain more deeply and are much smaller in gross size. These facts lead one to suspect that the cells of the duct have much less of the work of the gland to perform than do those of the alveolus. If we take as our conception of the work of a gland-cell, that it receives materials from the blood which it transforms chemically into some form of product, and which it throws out into the lumen thence to find its way into the mouth cavity, we readily see that it is equally suitable that the active alveolar cell should show a large front to the blood as its pyramidal shape admirably fits it to do, while the duct should expose an equally small area as it does by reason of its very small base and tall columnar form.



Artery in optical section, from parotid gland.

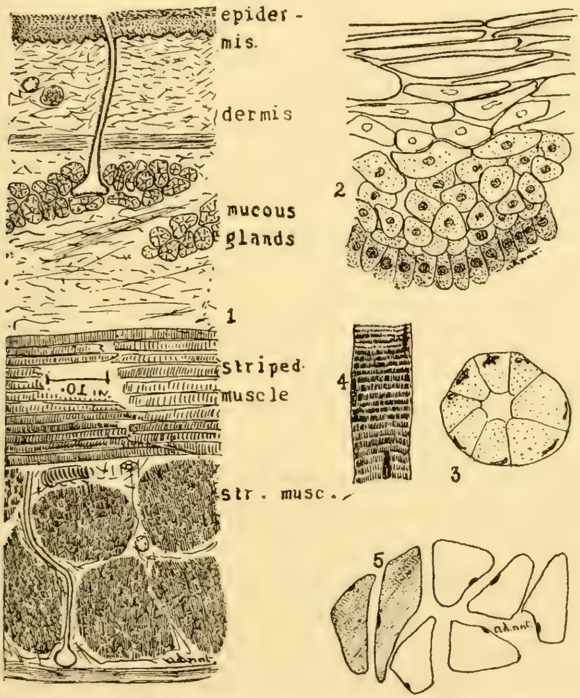
Sections of the salivary gland, as in fact of all the alimentary tissues, give good opportunities for this study of the structure of arterioles and capillaries. In studying the former, one of the smallest of the arterioles should be selected, in a place as free from other objects as possible. The microscope should be then carefully focussed, at one time on the surface, and at another on the centre of the tube, so as to obtain an optical section. In the surface view the unstriped muscle cells are visible clasping the tube, in some of these the cell is seen full length on the top of the tube, in others a part is seen on top, the rest being on the other side. In the centres of these cells the nucleus is clearly seen. The nucleus is recognizable by its deeper stain. Underneath the muscular coat can be seen other nuclei, which run in a transverse direction, these are the nuclei of the cells which form the inner

coat. In the optical section the edge of the tube appears to be bordered by cubical cells, this is the optical effect of the muscle cells, obtained by the view of their optical section. In the centre of some of these, the nucleus is seen, but others are not cut in its plane and hence do not how it. The capillary is seen as a thin parallel walled tube, with enlargements at regular intervals due to the position of the nuclei of the endothelium cells.

THE ŒSOPHAGUS, exhibits the three coats with great distinctness, on the surface it presents a layer of epidermis, in character like that on the tongue, but not thrown into papillæ; beneath the epidermis is the dermis, the vascular and connective tissue portion of the first coat, this is a compact mass of fibrous matter, penetrated here and there by smaller vessels and in places by the duct of the mucous glands which lie in the submucous coat. At the bottom of the layer is some muscular tissue. The middle coat is composed of loosely arranged connective tissue, in which the chief blood-vessels lie, in this layer also are situated the glands, which, known as the œsophagaeal glands, secrete the mucous secretion needed to moisten the surface of the organ, so as to reduce friction in swallowing. The vertical section accompanying this shows the duct leading down into the submucous coat, where it dilates to run to the various portions of the gland, no section will ever be found to show the full length of the duct, as this semi-diagrammatic drawing does. Fully half of the wall of the œsophagus is composed of muscular tissue, this is arranged in two portions one circular and one longitudinal, these when cut are seen, one cut lengthwise, the other, cross-wise, out side of the muscular coat is a denser layer of connective tissue forming a sheath for the tissues beneath.

The examination of the different parts of the section with the high power will demonstrate the following points, the epidermis is composed of cells which lie in several

layers and are different in different levels. The cells nearest the surface are very long and slender in vertical sections; if, in any part of the section, a surface view of these cells is given, they are there seen to be broad, these cells are destitute of any living granular protoplasm, though the shape of the nucleus is retained in the centre. Cells similar to these can be had by mounting a lit-



Vertical section of the œsophagus, with high-power view of the epidermis, gland, and muscular tissue.

tle saliva, in which they are found, having been scraped off the surface of the cheeks. Below the outermost layers the cells are not so greatly flattened though still considerably elongate and some slight traces of granular particles are visible internally, still lower the cells are rounded and their nucleus and cell-protoplasm are evidently alive, finally at the bottom of the layers is a sin-

gle layer of columnar living cells, which, by their absorptive affinity for the staining fluids, are plainly shown to be active living cells. These cells like epidermis generally, grow most rapidly on the lower layer, where they are nourished from the blood in the capillaries directly beneath them. Their multiplication results in crowding, on the lower level, which pushes up some while it leaves the rest. Some cement substance secreted by the cells causes them to cohere, and since they are less well nourished they degenerate in form and composition from the columnar through the rounded to the flattened shape, probably as a result of the pressure which is brought to bear on them from the outer surface. The removal of cells from the surface as a result of friction there, is made good by the fresh arrivals sent up from below, as a result of the multiplication which is constantly going on in the lower living layer.

The structure of the glands in the middle coat p. 143 is similar to that of the salivary gland, it is a typical racemose gland, the wedge shaped cells, which line the alveoli leave a central lumen into which their secretion can be drained into the duct and carried out to the surface where it is to be of service. The duct on the other hand is lined with cells whose shape and position does not adapt them especially for work as secreting agents, there is thus here a case of that division of labor and diversity of form which is so prominent a fact in biology.

The most of the alimentary canal is lined with a muscular coat composed of unstriped muscular tissue, the structure of which will be more particularly pointed out in connection with the stomach and intestine. Some of that tissue enters into the wall of the lower end of the gullet, but the most of the wall is composed of striped muscle. The striped muscle is used most commonly in the body, in the formation of the muscles which are employed to move the bones. The reason for this is that

this kind of muscle works much more rapidly, than does the unstriped muscle. From the fact that the striped muscle is used in the skeletal muscles, most of which are under the direct control of our will, it has often been called "voluntary" muscle. In the gullet, however, it is not at all under the control of the will and the act of swallowing is only voluntary in its initial stages. This kind of tissue is composed of "fibres" of great length and very slender, the fibres are not cells, but are built up out of cells. The exact way in which the cells are arranged and the fibre formed is not sufficiently understood, but we may say that the nuclei of the component cells are on the surface of the fibre where they are visible as elongate slender deeply staining masses, just below a thin and very delicate membrane, the "scarcolemma" which encloses the entire fibre. The nuclei of which there may be a great many in one fibre are enveloped by a network of protoplasmic fibres which reach out through the fibre in every direction. The mass of the fibre is composed of a chemical material called "myosin" which is physically very much like the white of an egg, that is, it is semi-fluid and coagulates on boiling or on exposure to acids. This substance of the fibre is placed in the fibre in such a way that the fibre, when examined microscopically, presents an appearance somewhat like the surface of a ribbed ribbon. This appearance, thus due to the way the myosin is arranged in the inside of the fibre, has given the name of striped muscle to this variety of muscular material. It seems to be probable that the myosin is produced and arranged in the fibre by the net work of protoplasm, if so then the fibre as we see it is mainly the production of the real, but unseen protoplasm. The views given in the cut on p. 143 show the ordinary appearance of the fibre in both longitudinal (4) and cross section (5).

THE STOMACH differs from the gullet in respect to both

the muscular and the mucous coats. The sub-mucous coat is the seat of the supply vessels and nerves in all parts of the alimentary canal, in the section of the small intestine, p. 149, is shown how the artery bores its way straight through the muscular coat, not at that time giving off any branches, but reserving them to be given off in both directions from trunks which lie in the submucous coat. The mucous coat is composed of tubular glands which are generally branched near the surface into three or four simple straight tubes running vertically to the surface of the coat. These tubes are so closely packed that at first it is very difficult to analyze the appearance of the section and to distinguish that in the confusion of cells there is any definite structural order. In an injected specimen the capillary arrangements are clearly displayed, but the grandular structure is equally difficult to recognize. The vertical section (see frontispiece) shows, somewhat diagrammatically, the actual mode of arrangement of the different parts of the mucous coat.

At "a" and "b" are the openings into the stomach of the ducts of the glands, in the depth of each of these there is a row of cells running down close beside others of the same sort, these, as seen in the one opening at "a" are in the form of a long and narrow tube with a central lumen, two tubes are shown here opening by a single duct. The high power shows the character of the cells in the walls of these tubes, these cells are different at different levels, at the summit they are tall and very slender with a long and narrow nucleus, the outer ends of the cells are rounded showing their soft character as in the free portions of cells in the liver. The cells on the free surface of the duct are vertical to the surface, and in the deeper parts of the duct they are again vertical, but between these points they are oblique, leaning upward, the cells are nearly transparent containing very little stain-

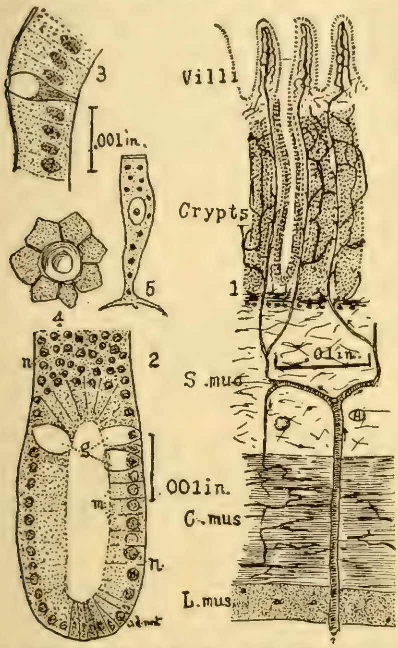
able matter, and thus in the mass look like a clear border in the section with a row of deeply colored spots, the nuclei running through them. In places directly underneath these cells of the uppermost level, in the midst of the fibres of areolar tissue, can be seen a section of one of the minute capillaries, and perchance, this may contain a few corpuscles, this is the constant position of the blood vessel as compared with the cavity of a secreting organ, which opens on the free surfaces of the body, namely with a layer of living cells coming between the blood and the cavity. The cells thus on the surface and in the duct portion of the tube are directly continuous with those which form the deeper portions of the gland, (figs. 2, 3,) the latter pass insensibly from the former, as seen at the bottom of the ducts in some places in the section. In the recess of the tube there are two kinds of cells, the cells which more directly line the tube are approximately four-sided in section, they nearly fill the lumen, their free ends being somewhat swollen, each has a large round nucleus which nearly fills the cell. Besides these cells that are characteristic of glands generally, the tubular glands of the stomach are in places nearly covered by cells called "peptic cells" or "cover cells," which have the shape aud relation shown in figs. 2, 3. These large flat cells lie on the deeper side of the ordinary glandular cells somewhat as the demi lune cells do in the salivary glands. In a section through the middle of the tube (fig. 3,) a peptic cell is seen to have a triangular shape, and to be wedged in between the other cells. These large cover cells are so conspicuous in most sections as to nearly or completely obscure the other cells.

The relation of the blood vessels to the tubes is shown in the general view, the capillaries arise from the horizontal vessels in the submucous coat and run up between the tubes, in this situation they surround the tubes, but never pierce through them to enter their cavities, so that

the deeper ends of the cells are in relation with the blood supply while their opposite ends open on the lumen. This is exactly the relation of cell and vessels in all glands, and it enables them to work to the best advantage, for the cells can attract from the blood those elements it shall use, and can throw off its products into the lumen whence they are removed and thus their accumulation is avoided. At the bottom of the layer there is a layer of muscular tissue, called the "muscularis mucosæ." The third coat of the organ is the usual muscular coats this is composed of unstriped muscle tissue, a fuller account of this tissue will be given and the cells illustrated in connection with the small intestine.

The small intestine, a tube in man thirty-nine feet long and an inch in diameter, has a similar structure throughout its entire length. The cut of vertical section p. 149, will show the structure with sufficient clearness for the purpose of interpreting sections made in an ordinary way. There are three coats, the two active coats, muscular and mucus, and the accessory coats, submucous, in which the vascular and nervous trunks lie, and from which they are distributed to the active layers. The mucous coat on the inside is composed of a layer of tubular glands, "crypts," as in the wall of the stomach, though the glands are not the same histologically as those of the stomach, and in addition of a layer of structures which are peculiar to this organ called "villi." A little reflection on the facts shown in the figure will show any one that a villus is the opposite of a tubular gland; a gland being a depression from the surface which is lined with cells, while a villus is a projection which is covered with cells. The difference is seen by a consideration of the position of the blood-capillaries in the two cases; in the first, the capillary surrounds the tube; in the second it is inside, see fig. 1, p. 149. From the point of view of the cell the relation is unchanged for

the blood is still on one side and the cavity on the other, but as a result of the difference of position, the villus is adapted to the absorption of products which surround it in the chyle, while the gland is suited rather for the work of secretion. There is a considerable difference in the histological character of the cells from different parts of the mucous coat.



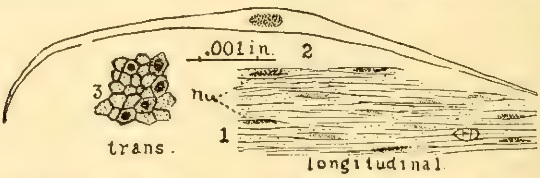
Vertical section of the wall of the small intestine, with details of the cells of the villus (3 and 4) and of the glands (2). One cell (5) is filled with engulphed fat droplets.

Fig. 4 shows a section in the depth of the gland, its lumen is large, there are no cover cells, and the gland cells are higher than broad, at the upper part of the drawing, a surface view of the tube is represented, where the section passes out of the plane of the lumen.

Some of the cells of the gland are swollen and empty, these are not so numerous as in the crypts of the large intestine, they are called "goblet cells." They are usually supposed to be mucous cells which have discharged their contents. The cells of the villus are shown in figs. 3, 4, and 5. In the first a small part of the villus with eight adjoining cells are shown, one of these is a goblet cell, in 5 a single cell isolated from the rest by maceration, in an animal killed soon after a meal containing a large amount of fat. The fat drops have been demonstrated by staining with osmic acid, and are seen to be inside the cell. In fig. 4, a surface view showing a goblet cell and several surrounding cells of the villus is given. The

cells are thus shown not to be square in cross-section as might be imagined, but five or six sided. They are the form that would be produced on nearly equal yielding cylinders, placed side, by mutual pressure.

The outer coat of the small intestine is one of the best objects for the study of unstriped muscle tissue. In sections of the organ the mode of association of the cells can be determined after the separate fibres have been isolated and examined in specimens which have first been macerated. The maceration in alcohol 30 per cent, 24 hours is sufficient for the purpose of separating the cells.



Unstriped muscular tissue, Small Intestine.

Figure 1 of the accompanying cut is in a single cell accurately drawn with the camera lucida from such a preparation. It is extremely long and slender, with a central nucleus. Figures 2 and 3 are from the muscular coats in an ordinary section of the small intestine, in the former the nuclei are visible and the general parallelism of the cells but a wrong idea of the length of the cells would be derived from such an examination because the ends are so slender that they are lost in the mass, the two figures are drawn on exactly the same scale, and the nuclei if compared are seen to be alike in size. In a cross-section the cells appear polygonal in outline, and their nucleus is seen to occupy the centre of the cell, the reason that no nucleus is seen in some of the cells as well as that they are not all of the same size is because all are not cut in the same level of the cell.

The pancreas is closely similar to a salivary gland. In general sections the ducts and their divisions are shown as in the parotid, and the mass is composed of

the same closed alveoli. The alveoli of the organ, however, are not so often circular in cross-section but are more elongate, so that the organ is less strictly a racemose gland than is the parotid. The section illustrated is drawn to show the different appearance of the gland alveoli at different hours of the day, for in the pancreas there is a marked difference in the shapes of the cells, this depending on the time after the animal partook of its last meal, when it was killed. In the case of the active organ during the period of digestion, while being compelled through the nerves to furnish its product to the intestine, the cells are shrunken so that the central lumen is clearly visible and the cell-walls themselves are also evident.


1

Pancreas, four hours after eating. Cat.


2


3

A few hours after the act of digestion while the organ is having a chance to partake, like the cells of the body at large, of the results of digestive work, the cells of the organ fill themselves with their peculiar material, out of which they can construct trypsin, their specific secretion, this they do in preparation for the next meal. The cells in an organ of an animal killed at such a time are so far swollen by the accumulation of this material that the cell boundaries are wholly or nearly obliterated while the lumen is also seen with difficulty if at all. It is because the animals are so likely to be killed in this condition that as a rule sections of the pancreas exhibit cells so indistinctly. Figure 1 of the cut was drawn from nature, from a pancreas of cat four hours after a full meal. Figure 2 is from a "resting pancreas" and

fig. 3 is from an "active one," both are copied from the standard text-books for comparison with the fig. 1.

The large intestine is a tubular organ of larger diameter than the small intestine, it presents the usual three coats in the usual order. The mucous coat is the only one which requires special mention, that is composed of broad and short tubular glands, much like those of the small intestine which are called the "crypts of Lieberkuehn." There are no villi. A student who has mastered the structure of the stomach and small intestine will find it an easy matter to interpret his sections of the large intestine.

Biological Laboratory of Hamline University.
Feb. 17, 1894.

The Aniline Stains.

BY SMITH ELY JELLIFFE, M. D.
BROOKLYN, N. Y.

The use of the aniline stains for histological and pathological work for both plant and animal tissue is daily increasing, and almost as soon as commercial enterprise has evolved a new dye stuff for domestic use, the workers in microscopical lines have seized upon it and the results are soon found in the various journals.

There has been or are still a number of difficulties in the way of procuring these stains, and many times the endeavor of the microscopist to corroborate the results of other investigations fails. The reasons are not far to find: Many times it is the unscrupulous clerk who pencils upon the bottle Acid Fuchsin, when it should be Fuchsin; or Methylen Blue, when the original is Methyl Blue; and unless the investigator is aware of the differences the results are not satisfactory. Again it is the fault of the original investigator who describes the use of Orange, in watery solution, not mindful of the fact that there are ten to twenty Orange preparations in the market.

The writer has encountered these obstacles and many more within the past few years in histological and pathological work and has considered that a collection of some of the more important stains should be made; a list of these synonyns, their appearance and some of the tests, to determine sophistication. The names and tests are drawn in the main from Beilstein's Handbuch der Organischen Chemie, and the Tabellansche Ubersicht der Kunstlichen Organischen Farbstoffe, von Gustav Schultz und Paul Julius: Zwirte Auflage: Berlin, 1891, upon whose "Alle Rechte vorbehalten," I hope not to have infringed.

The order of description in tho main is: The name of the preparation which is more commonly used, its synonyms, the appearance of the dye in the rough, solubility in water and alcohol and its behaviour with acids, Hydrochloric (HCl), and Sulphuric (H_2SO_4), and with alkalies, Sodic Hydrate (NaOH).

ALKALI BLUE. Nicholson Blue. Soluble Blue. *Soluble Aniline Blue.*

A light or dark blue powder.

Difficulty soluble in cold; more readily soluble in warm water. In alcohol somewhat soluble. HCl to watery solution, blue precipitate. NaOH to watery solution, red brown solution. Concentrated H_2SO_4 soon brownish red. Dilution with water, blue.

AURANTIA. Kaiser yellow.

Reddish brown crystals.

In water, soluble with orange yellow color.

BISMARK BROWN. Manchester Brown. Phenylen Brown. Vesuvin. Aniline Brown. Canelle. English Brown.

Dark brown powder.

In water soluble with brown color. HCl to watery solution, no change. NaOH to watery solution, brownish precipitate. H_2SO_4 Conc. brown solution. Dilution with water, red.

DAHLIA. Hofmann's Violet. Iodine Violet. Primula. Red Violet 5 R. extra. Violet 5 R. Violet R. Violet RR.

Two qualities (a) Reddish and (b) Violet.

(A). Green crystalline powder. In water soluble with fuchsin red color. HCl to watery solution, yellowish brown color. NaOH to watery solution, brown precipitate. H_2SO_4 Conc. yellowish brown solution. Dilution with water, no change.

(B). Shining green pieces. In water, easily soluble with blue violet color. HCl to watery solution, first green then yellow. NaOH to a watery solution brownish red precipitate. H_2SO_4 Conc. brownish yellow solution. Dilution with water, at first olive green, then green, then blue.

CYANIN. Chinolin Blue.

Green shining crystals.

In water, insoluble in cold ; by heating with difficulty soluble with a violet blue color. HCl to a watery solution, decolorization. NaOH to a watery solution ; cold, a bluish bronze like precipitate; warm, a brownish precipitate. $H_2\ SO_4$ concentrated, colorless. Dilution with water colorless.

EOSIN. Eosin A. Eosin GGF. Water Soluble Eosin. Eosin B.

Reddish, blue shining crystals or brownish powder.

In water, easily soluble with bluish red color ; the diluted solution shows a greenish fluorescence. In alcohol readily soluble with bluish red color and yellowish green fluorescence. HCl to watery solution, yellowish red floculi. NaOH to watery solution, no change. $H_2\ SO_4$ Conc. yellowish solution. Dilution with water, yellowish red precipitate.

FUCHSIN. Rubin. Magenta. Aniline Red. Rosein.

Found as Chlorhydrate, Acetate, Nitrate and Sulphate.

As Chlorhydrate, Cantharides like shining crystals. As Acetate, irregular green shining pieces. As Sulphate, fine green shining crystalline powder.

In water, soluble with red color. In alcohol soluble with red color. HCl to watery solution, yellowish. NaOH to watery solution, decolorization. $H_2\ SO_4$ Conc. yellowish brown solution. Dilution with water, nearly colorless.

FUCHSIN, S. Acid Fuchsin. Rubin S. Acid Rubin. Acid Magenta.

Metallic green shining grains or powder.

In water, soluble with bluish red color. In alcohol insoluble. HCl to watery solution, no change. NaOH to watery solution, complete decolorization. $H_2\ SO_4$ Conc. yellowish solution. Dilution with water, gradually red.

INDULIN. Indulin N N. *Nigrosin* sol. in water. True Blue R. True Blue 3 R.

Indulin, bronze shining powder. Nigrosin, black shining fragments.

In water, soluble with a blue violet color. In alcohol, blue solution. HCl to watery solution, becomes more blue. NaOH to watery solution, brownish violet precipitate. $H_2\ SO_4$ Conc. blue solution. Dilution with water, violet solution.

IODINE GREEN. Night Green. Metternichs Green. Vert lumiere.

Hard dark green pieces.

In water, easily soluble with a bluish green color. HCl to watery solution, reddish yellow color. NaOH to watery solution, colorless. $H_2\ SO_4$ Conc. soluble with reddish yellow color. Dilution with water, weak yellowish green.

METHYL BLUE. Brilliant Cotton Blue. Methyl water Blue.

A dark blue powder.

In water, soluble with a blue color. H Cl to a watery solution, no change. Na OH to a watery solution, a reddish brown solution. $H_2\ SO_4$ Conc. soluble with red brown color. Dilution with water, a blue solution.

METHYL GREEN. Paris Green. Vert Etincelle. Light Green. Green of Methyl Aniline. Double Green. Green Powder.

Green Crystals.

In water, easily soluble with bluish green color. Insoluble in Amyl alcohol. H Cl to watery solution, yellowish red color, which becomes yellowish green on addition of water. Na OH to watery solution, becomes colorless. $H^2\ SO_4$ Concentrated soluble with yellowish red color. Dilution with water, yellowish green solution.

METHYL VIOLET B. Paris Violet. Direct Violet. Violet of Methyl Aniline. PYOKTANIN.

Metallic green shining fragments or powder.

In water, soluble with violet color. Alcohol, soluble, also in amyl alcohol. H Cl to watery solution, at first green then on addition of more acid, deep golden brown solution. Na OH to watery solution, brownish red color and precipitate. H_2 SO_4 Conc. yellow. Dilution with water, yellowish green, then bluish green, finally violet.

There is another Methyl Violet. 6 B. Benzyl Violet. Paris Violet. 6 B. that comes in metallic brownish shining fragments or powder. Its reactions are identical with Methyl Violet B.

METHYLEN BLUE. B. B.G. B. B. in powder extra D. Methylen Blue. B. B. in powder extra. Aethylen Blue.

Dark blue or reddish brown, bronze shining powder.

In water, easily soluble with blue color. In alcohol soluble. HCl to watery solution no change. Na OH to watery solution, violet color. Much concentrated Na OH produces a dirty violet precipitate. H_2 SO_4 concentrated golden green solution. Dilution with water, blue solution.

PICRIC ACID.

Pale yellowish crystals.

In water, with difficulty soluble in cold, more readily soluble in warm water. Soluble in alcohol. Benzol : melts at 122.5 and has a bitter taste. With Potass. Cyanide, a brown solution.

SAFRANIN. Safranin T. Safranin Extra G. Safranin S. Aniline Rose. Old name Pink.

Red brown powder.

In water, soluble with red color. In alcohol, red solution with yellowish red fluorescence. HCl to a watery solution, blue violet solution. NaOH to watery solution, brownish red precipitate. Conc. H_2 SO_4 green solution. Dilution with water, from blue passing to red.

EDITORIAL.

Progress.—When the present President of the Royal College of Physicians of London was a student at University College Hospital (45 years ago) one of the professors said to him : "There has been a novelty introduced into the wards. There is a microscope for ward A and one for ward B. Now if anything is so small that it needs a microscope to see it, I do not think it can be of very great importance."

To-day only an ignoramus of the worst sort or an insane man could make such a remark. The greatest sanitary battle to-day is with the unseen enemies of our bodies.

Meeting of Michigan State Board of Health, Lansing, April 13, 1894.—During the past year this Board has done much good work, including that in connection with quarantine and the prevention of the introduction of dangerous commun-

icable diseases. The Board has entered upon a most important work for the prevention and restriction of tuberculosis in man, and it is believed that the results will be great. This Board has taken the lead of others in declaring consumption to be dangerous to the public health, and has recommended advanced measures for its restriction.

Last year a useful Conference was held with special reference to cholera. That subject is still of interest and dangerous immigrants are still coming into Michigan. But it is proposed this year to give special attention to that disease which is already here and causes most deaths—consumption, and to give the health officers opportunity to study the subject at the State Laboratory of Hygiene where the bacteriological and other facts relative to the causation of this disease can be so well demonstrated.

MICROSCOPICAL APPARATUS.

A New 1-5 Objective.—In examining an object while still in the compressor, such as I described in the last number of this Journal, having glasses of 1-10 inch in thickness, one is naturally restricted to objectives of low power. In my special work, however, it was quite important that a higher power should be occasionally used, and I made inquiry with negative results of all the leading makers, for a 1-5 that would work through a 1-10 inch cover. I found that such an objective was not on the market, but Messrs. Spencer and Smith of Buffalo, N. Y., offered to design and make such an objective, and having received permission from the U. S. Bureau of Animal Industry to give the order, I received later on an objective, which was claimed by the maker to give the results required.

I find this 1-5 objective of 50° air angle, has very fine definition, the correction for the abnormally thick cover being perfect, but it will not work with ordinary glass covers, it being impossible to make such a lens without collar adjustment.

The use of glasses 1-10 of an inch thick, allowing great pressure to be made, and sustained in a compressor frame, having a screw adjustment at each end, has a wide field of usefulness, and as the object can now be examined while under this compression with as high a power as one-fifth, it opens new and

useful opportunities not hitherto possessed by the microscopists, of special value to the Botanist and those making biological researches.

JOHN MICHELS.
Boston, Mass.

MEDICAL MICROSCOPY.

Work of the Mich. Board of Health in 1894.—The subject of tuberculosis in animals as a cause of tubercular diseases in man, is being presented to the people. A survey of the cattle and milk in different parts of this State will show in what way and to what extent the health and lives of the people are endangered by tuberculous meat and milk. Dr Gray reports that he has conferred with veterinary surgeons in Pontiac and Detroit, has visited the health department in Detroit, that he has corresponded on this subject with the three members of the State Live Stock Commission. Hon. J. J. Woodman has said that the presence of tuberculosis in animals is not being reported to the State Live Stock Commission.

Dr. Baker says he has conferred with the State Veterinarian, who advised further and personal conference by this Board with the State Live Stock Commission. Dr. Baker quotes from the last report of the commission relative to tuberculosis—"It is beyond question both infectious and contagious, particularly in the pulmonary developement or consumption of the lungs." * * * "Years of added experience and careful observation lead us to the conclusion that the annual losses among Michigan cattle from tuberculosis are much greater than from all the other contagious diseases affecting our domestic animals, and that the disease is steadily increasing. We have given the subject very careful thought and consideration, and have as yet failed to find a satisfactory plan for its treatment or extermination." * * * "It, as yet, is one of the unsolved problems, lying all in front and like some bridges, in our pathway, the day is not far distant when an attempt must be made to cross." Secretary Baker thought that now is the time to make the attempt to "cross the bridge," and earnestly hoped that the State Live Stock Commission would co-operate in the effort for the restriction and prevention of tuberculosis in animals and in man. It was voted that the President be requested to call a special meeting of this State Board of Health at such time as arrangements can be made, for a joint meeting with the State Live Stock Commission, to consider the subject of the restriction of tuberculosis in animals and in man.

As chairman of the Standing Committee on "Epidemic and Communicable Diseases," Prof. Vaughan made a report on the subject of the restriction of tuberculosis in man by means of a proposed State Hospital for Consumptives, this subject having

been referred to him, at the last special meeting. The subject was discussed at great length, and resolutions were adopted as follows:

Resolved, That we recognize the following facts:

(1). That tuberculosis is the most grave and fatal disease now affecting the health and lives of the people of this State, destroying about 3,000 lives per year.

(2). That this disease originates principally by transmission from man to man or from man to animals and again to man.

(3). That the spread of this disease can be best arrested by the disinfection of the sputa and other discharges, by special supervision of those infected, and by the care of such persons under conditions which will prevent the transmission of the disease to others.

(4). That such disinfection and supervision can not be carried out in the crowded homes of the poorer classes, and

(5). That under conditions which will prevent re-infection, many consumptives may be permanently cured, and returned to their homes and work, educated in the methods of restricting the disease. In view of these facts,

Resolved, That this Board request of the next Legislature an appropriation of $ for the purpose of building, equipping and maintaining a State Hospital for Consumptives.

Prof. Delos Fall presented a preamble and resolution which were adopted as follows:

Whereas It is desirable that every step taken shall tend toward giving the largest amount of sanitary education to the teachers and to the people of the State therefore.

Resolved. That it is the judgment of this Board that the proposed State Hospital for Consumptives should be located at the seat of the State University at Ann Arbor, in order that it may afford the best opportunities for the observation and study of this most important disease, in conjunction with the investigations now being so satisfactorily pursued, in bacteriology and other departments of sanitary science, at the State Laboratory of Hygiene.

The Secretary presented and read portions of his report of work done in the office during the quarter just ended, which included the action taken for the restriction of 412 outbreaks of dangerous communicable diseases, 6 outbreaks being of small pox. Consumption was reported from 185 localities.

BACTERIOLOGY.

Mice Destroyed by Bacilli.—Jean Danysz reports using a specific bacillus, for destroying immense numbers of small rodents in France. He does not tell what the bacillus is, but he

issolated it by the culture method. The contents of 125 gelatin culture tubes were dissolved in 50 litres of water. In this he soaked 80,000 cubes of bread each about 1 c.c. The bread was scattered through the 75 hectares infested with 10,000 to 30,000 mice. The total cost was very slight. Within 3 days after the scattering of the bread sick and dying mice were to be seen on all sides. The microbes were found in their blood. After 15 days scarcely a mouse could be seen alive and in their burrows great numbers of carcasses were present. The microbe is said to be similar to that of duck cholera, but fowls are not susceptible to it. Cats, dogs, fowls and domestic animals are not injured by it. It is proposed to try this method on the gophers which destroy so much grain in the West.

Gen. Hawley on Bacteria.—Gen. Hawley in a speech on the Coxey movement and in reply to Senator Allen of Nebraska said that the latter's speech: "Had in it, not requiring a microscope, but visible to the naked eye, the bacteria and bacilli of anarchy."

MICROSCOPICAL SOCIETIES.

Microscope Club, Lincoln, Nebr.

March 28.—In the absence of the president, Dr. Ward presided. On recommendation of the auditing committee, the report of the treasurer was adopted. Prof Seawell exhibited sections of a lamprey by S. H. Gage. Dr. Bessey exhibited a set of sections of the leaves of conifers. Mr. Dales had sections of rotten pine wood, showing large numbers of spores of what seemed to be a slime mould filling all the interior. Dr. Philbrick showed sections of a "sago" spleen; and Dr. Ward exhibited some fish parasites (*Distoma*).

NEW PUBLICATIONS.

Mosses.—The book with which to study mosses is Jameson's Illustrated Guide to British Mosses, for sale by the author, H. G. Jameson, 6 College Road. Eastbourne, England. It contains 59 plates, or 2400 figures, descriptions of the genera but not of the species.

The Refractionist.—This is the name of a new monthly

magazine devoted to Ophthalmology. Price $2.00—same size as THE MICROSCOPE (price $1.00), yet it may die for lack of support inside two years. We hope not but we think that specialization is progressing faster than the means of support.

Practical Methods in Microscopy. By Charles H. Clark. Boston, D. C. Heath & Co., 1894. 12° pp. 233. 41 cuts, 17 plates.

Believing that none of the existing volumes on the microscope were sufficiently concise and explicit for the beginner, and for those who have no teacher, the author has undertaken to briefly present all that a beginner needs to know in order to use the microscope successfully. Gage's book is for the student under guidance of a teacher; Clark's, for the solitary student. He certainly has touched upon a great number of things of elementary importance and has made very plain what others sometimes leave unexplained assuming that their readers "know something." The apparatus of Messrs. Bausch & Lomb is quite fully pictured and described, and this makes the book of more value in America than that of Cross & Cole, who describe English apparatus only. Bacteria, photomicrography, polarization, and other collateral topics are briefly summarized.

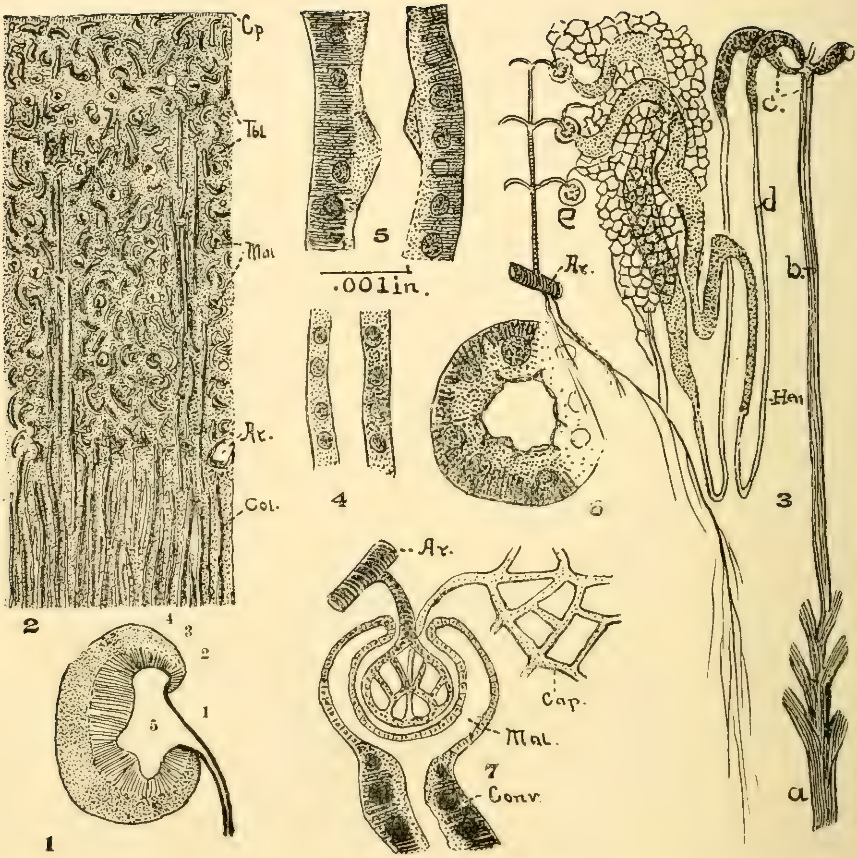
Kindness to Animals. A. S. P. C. A. New York, 1893, 24° 53pp.

This little catechism of 398 questions and answers is, we are astonished to find quite sensible- Nearly everything said and done by societies for prevention of cruelty smacks of crankism, silliness and folly, but this book, though containing a few foolish things, is pretty good. The fact that the English sparrow is an intolerable pest is ignored and children are encouraged to throw crumbs to it. Boys are warned that "it is cruel to catch fish not good for food" (i. e. good for oil, guano, etc) but we ask is not the cruelty the same in all cases ?

An earnest effort should be made to introduce such a book into Sunday School classes.

An Introduction to Structural Botany. By Dukinfield Henry Scott. London and New York. Macmillan & Co. 12 mo. pp. 288, figures 113. Price $1.00

To the microscopist the book is of interest as explaining fully what is to be seen in plant life by means of the microscope though the author alludes to the microscope only incidentally.



HISTOLOGY OF THE KIDNEY.

THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. JUNE, 1894. No. 6.

Studies of the Histology of Various Mammalian Tissues.---III.

BY HENRY L. OSBORN,

ST. PAUL, MINN.

Continued from page 135.

WITH FRONTISPIECE.

ON THE CELL STRUCTURE OF THE MAMMALIAN KIDNEY.

There is hardly any organ the histological structure of which is more confusing and inextricable on a first study than the kidney. This is due, in part, to the large number of different tissue elements which enter into its composition, and in part to its tubular structure, in consequence of which the different parts of the same tube will be separated in sections and have no apparent relation. Besides having a good section properly preserved

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EXPLANATION OF PLATE.

Fig. 1. Diagram of median section through the entire kidney, 1, ureter ; 2, malpighian pyramids ; 3, cortex ; 4, capsule ; 5, pelvis

Fig. 2. Low power view of the cortex and a small part of the malpighian layer of the kidney. The tubules are slightly less crowded than in nature, for the sake of clearness.

Fig. 3. Diagram to show the different parts of a renal tubule, a, collecting part ; b, collecting tubule ; c, branchings of collecting tubules ; d, the loop of Henle ; e, the convoluted part and the terminal "malpighian body."

Fig. 4. A small part of a loop of Henle in median section. Camera lucida, x500 diam. Same as fig. 5.

Fig. 5. Convoluted tubule showing the cells in section, the lumen in the centre and the rodded structure in the basal ends of the cells. Cam. Luc., x500 (nearly). See scale.

Fig. 6. Cross-section of convoluted tubule.

Fig. 7. Termination of tubule showing the location of the vascular supply, and the epithelium of the tube and of the cup, somewhat diagrammatic.

to show the cell structure, and another one injected to show the position of the vascular system, the student must bring some degree of experience to bear on the study before he can hope to interpret the real structure of the sections which he may have before his eyes. If he has, by study of such glands as those already described, the salivary glands, the mucous coats of the alimentary tube, and such comparatively easy subjects, learned to recognize ducts, secreting parts of glands, capillaries, arterioles, lumina, basement layers and cell boundaries, he has some reason to expect to understand the kidney. If he has not had this preparation, he will find the kidney a poor subject on which to make his first acquaintance of gland histology.

I shall suppose that the reader of this paper has read the preceeding numbers of this series and that the parts of a gland are intelligible to him. I shall also suppose an acquaintance with the mere anatomical facts about the organ such as are given in any elementary book of anatomy. That is, to say in brief, that there are two kidneys, concavo-convex in form, that each receives a branch "renal-artery" from the aorta and sends a vessel, the "renal vein," to the inferior vena-cava. That each also gives off a duct, the "ureter," which runs direct to the urinary bladder. I shall also assume the coarse structure of the organ (fig. 1), its hollow pelvis, which is an expansion of the ureter; the cortical portion, made up of the vascular structures and of the tubules, these running radially from the pelvis to the surface, and I shall attempt to picture, in detail, the structure of the cells of the tubule and the situation of the vascular supply, in such a way as to enable one possessing the necessary sections to find these parts for himself.

A word in regard to the preparation of sections will not be amiss. Injections of the kidney can be made by inserting the cannula in the dorsal aorta and forcing the

fluid down, after ligaturing the aorta below the renal artery. This will force the fluid into the arterial system and around and out by the way of veins. Material which is to be hardened for sections of the cells should be cut with a very sharp blade, from a kidney taken from a very recently killed animal. The pieces should be cut in the form of wedges not more than a quarter to half an inch thick at the base. These should be hardened in an abundance of the hardening reagent. Sections should be extremely thin; as the tubules are not much more than a thousandth of an inch in diameter, it is obvious that the sections will not reveal their interior lumen, if they are more than that thickness.

The first thing to do is to "orient" the section, that is to say, to determine its position in the organ from which it came. Very often a student will neglect to orient his section in the beginning, and thus will obscure all his subsequent work. It is possible to keep memoranda of the position of the piece, if you cut your own section, from the time it comes out of the organ until the time it is mounted on the slide. In the case of the sections made by students of embryology this must be done or the future determination of the structure of the animal, or part of an animal, is rendered dubious to a certain degree. If this has not been done in the case of the kidney, reference to a description of the organ will enable you to place the section in its proper position. Figure 1 is drawn for this end. It shows diagrammatically the pelvis of the kidney, (1), around that the "malpighian pyramids" (2), and around them the "cortex" (3), the whole covered with the "capsule" (4). The dotted line in figure 1 shows the location of figure 2 which is a low power view also somewhat diagrammatic of the cortex and part of the malpighian pyramid. The remaining high power views are from the cortical portion of the organ which is alike in all parts.

In the low power view the capsules should be distinguished and then the at first confused mass of the cortex should be studied and the winding tubules and occasional circular "malpighian corpuscles" be distinguished. Then the tubular composition of the "malpighian pyramids" should be made out. These parts should be, all of them, accurately seen and located and their relation to the gross organ *realized* before anything more is done. I have italicized that word realized; I mean it literally. The reason that people do not have more intelligent notions regarding the animal body is because they do not *realize* the facts of cell structure, these facts being mere verbal expressions, living only on the pages of Quain's Anatomy, and not realities existing in the animal body. As the realization of the coarser histology, as shown by the low power, is completed, the higher power should be used in various parts of the cortical layer to demonstrate its detail. The beginner should not expect to see all the points in his section at a glance. Prolonged seach and careful, thoughtful attention at different sittings are necessary to the full exploration of a section; this is true of any organ and especially of as difficult an organ as the kidney. Many students expect a section to be as "clear as a figure in a book;" they are doomed to disappointment. If they stop here they are not of the stuff to make histologists and may as well either drop histology or change their expectations. A section, no matter how good it is, cannot reveal itself to a first glance of even an expert and much less to the first glance of a beginner. When I say then that a certain structure can be seen, I mean by the student who complies with these conditions.

The cortex is composed of three different elements all of which are more or less visible—they are the areolar tissue, the vascular tissues and the tubules. Let us begin with the last, as these are the easiest to demonstrate.

The tubules are long and narrow tubes which do not run straight but in a wavy course, and which are longer than the diameter of the cortex. The different parts of the tube are different in their cell structure. It is by the difference of cell structure that we shall at first recognize the various parts of the tubule; but the true way is by tracing with infinite pains the connections between all the different parts by the study of many sections and many parts of sections. In fig. 3, a diagram of a tubule is given. This shows that the tubules are not simple but are branched, several uniting to form the terminal tube. This end of the tube (a) lies in the malpighian pyramid; it is called the "collecting tubule." It runs out towards the cortex and finally branches into a number of portions from each of which a slender tube (d) runs down and part way back. This is called the "loop of Henle."

The loop of Henle is followed by a thicker tubular portion (e), the "convoluted tubule," this runs into the final part of the tube, the "malpighian corpuscle" (e). This entire tube is one continuous organ made of epithelium cells, one layer deep, with a central lumen and surrounded with blood vessels; it has thus the typical structure of a tubular gland. Let us attempt to get a clear notion of the epithelium in each of the different parts. In actually distinguishing these points on sections it is necessary to remember that many of the parts of tubule will be cut so as to present surface views, and such will appear entirely different from sections passing through the centres of tubules. In the collecting portion of the tubule the cells are cubical and clear and are largely nucleated. The boundaries of the cells in this region are more clearly seen. The cells do not, however, have the columnar character which is seen in the cells lining the ducts of such glands as the salivary glands and the tubules of the stomach. The loops of Henle are

lined with very flat cells. There are some of them seen in figure 4 which is a camera lucida drawing from nature. The cells are but little wider than the nucleus; their side walls are inconspicuous or invisible; they are in some cases even, contracted between the nuclei to less than their diameter. In the convoluted portion of the tubule the epithelium is very peculiar; indeed, none like it is to be seen in any other part of the body. The cells of this part are shown in figure 5, in longitudinal section; and, in figure 6, in cross section. They have a large nucleus and are densely filled with protoplasm. Their basement membrane is very distinct; the lumen takes a zig-zag course owing to the different heights attained by different cells. At the basal ends the cells present a peculiar appearance, called "rodded," from the rod-like marks or striations that are found there. The cells of this part of the organ are immediately succeeded by the cells of the "malpighian corpuscle."

This is one of the most curious structures in the range of histology. In the actual section, it is only possible to see the structure in occasional and very favorable cases by the study of which a figure, No. 3, is constructed. It is seen to be a bulb with a double wall such that the bulb has two cavities one which does not open into the lumen of the tubule; and a second, between the two epithelial layers, which does open into the lumen. This bulb is lined with small cubical cells.

It is clear from this account of the kidney that the epithelium is the active part of the organ and that its function is some form of secretion. It is not so easy to state clearly the different functions of the different parts of the tubules which are implied by the difference in the epithelium. Indeed this cannot be done with any degree of confidence. How much mere filtration is an important part of the process, or in how far true metabolism is the mode of action, and whether the "malpig-

hian body" or the convoluted portion or the loops of Henle are secreting or filtering parts, or are both, are all of them unsettled questions. The peculiar "rodded" structure of the cells of the convoluted tubule also calls in vain for a satisfactory physiological explanation.

It, remains, the problem of renal physiology, among many other questions, to isolate the various elements and to state clearly the respective functions of each, and the conditions which govern it in the working body. In view of the complexity of such a problem it is no wonder that physiology is still in its infancy.

It now remains for us to investigate the vascular and supporting structure of the kidney. This will be facilitated if the student has injected material from which to study.

The main divisions of the renal artery enter, as already noted, at the same place where the main renal duct, the "ureter," leaves the organ. These branches pass beside the pelvis of the organ, and, arriving at the "malpighain pyramids," run radially beside the collecting tubules to the level of the cortex where they branch sending arterioles each way, both toward the centre in the "malpighian pyramids" and out into the cortex. All the capillaries from these as in all glands ramify between the secreting regions of the organ. In this instance, capillaries from the arteriole run into the "malpighian corpuscles" where they form a capillary network and then a single vein emerges. Both the entrance and exit of these vessels is through the opening of the bulb which leads to the first cavity, already shown not to be in relation with the cavity of the tubule. Having come out of the bulb, the vein breaks up again into a second set of capillaries which surround in minute meshes the convoluted tubules and then collect again to run off, by way of the "malpighian pyramids," to the renal vein. The distribution of the blood is very clearly shown in figure

3, which is adapted from classical figures of the older German and French histologists.

In non-injected sections of the kidney, many of the facts just described can be shown, but not so readily. The same criteria as those mentioned in the study of the alimentary mucous tissues for the detection of arterioles and capillaries can be applied and the relation of these to the parts of the tubule studied out and thus the description and figures can be verified. Of the connective tissues nothing extended need be said. The outer capsule is a membrane of white-fibrous tissue, and between the tubules there are fine strands of areolar tissue. These do not form a system of optical importance and are visible only as fine threads or wavy lines.

BIOLOGICAL LABORATORY OF HAMLINE UNIVERSITY.
May 17, 1894.

How to Find Diatomaceous Earth.

BY WM. A. TERRY.
BRISTOL, CONN.

One of the most obvious places to look over for diatomaceous earth is the bank of a railroad cutting. Where such cutting has gone through a deposit it forms an ideal opportunity for examination, as each stratum can be separately explored with ease, and the whole deposit thoroughly studied. The lines of stratification are generally obvious in a fresh cutting—in all cases originally horizontal. A fresh-water deposit will show the overlying black stratum of muck or peat, and under this the characteristic ash-colored layer containing the diatoms.

Where a railroad embankment crosses a marsh or a body of water, the soft deposit is frequently forced up on each side, rising above the water to the height of several feet; by digging down from the apex of these elevations, a diatomaceous deposit can almost always be

found, generally at a depth of from four to eight feet. In digging for foundations of abutments of bridges or culverts, especially in salt marshes, diatomaceous earth is almost invariably thrown up, and, as I have before mentioned, ditches dug for drainage will often give opportunity for interesting finds. All these, however, belong to the chapter of happy accidents, and should not be allowed to limit the operations of the collector. The great majority of diatomaceous deposits lie buried and will never be brought to light except by explorations undertaken for that purpose. Under nearly every large swamp whose level surface shows that it occupies the bed of a former lake or pond lies a deposit of diatoms; in very dry seasons some of these may be reached by digging, but in many cases the water makes this difficult and some kind of boring apparatus will be useful.

I have a cheap home-made contrivance that answers the purpose in most cases. It consists of a cylinder of galvanized iron about four inches in diameter and some eighteen inches long, slightly flaring toward the top; the lower edge is sharpened and provided with an internal lip on one side, one inch wide and three inches long, inclined spirally upwards. This is to prevent the enclosed core of earth from slipping out when it is drawn up through the water, and it also helps in penetrating through the deposit. On opposite sides at the top are riveted stout flat strips of iron which extend upward to a socket in which fits the handle, keyed in so as to be easily removed. The handle is a stout pole of the necesary length, with a cross bar at the top.

In using this I first investigate the swamp and decide which were the deeper parts of the former lake, then I dig down with a spade as far as as easily practicable; then I commence boring. As soon as the cylinder is filled I draw it up and empty it out at the top and examine carefully. As soon as the black muck changes to a

brown deposit, I consider that I have reached the ancient deposit laid down in open water. Here I examine for diatoms, as from this point to the bottom where gravel or hardpan is found the deposit should contain more or less diatoms ; near the bottom the stratum should be quite rich.

In many swamps this stratum will be found to average about two feet in thickness, in others the strata may alternate to a considerable depth ; in those connected in a series at different levels through which a stream flows or did flow in ancient times the deposit may be of great thickness. I have known it to exceed seventy feet. These thick deposits are not necessarily any more ancient than others of moderate depth, the differing surroundings and circumstances being sufficient to account for them.

The boring apparatus described is intended for use only in soft deposits of no unusual depth, it brings up a large amount of material and will work under water, but for hard or dry deposits its use would be impracticable ; dry deposits are best reached by simply digging.

For the marine deposits of the salt marshes a cylinder two inches in diameter would probably be best. On the Connecticut shore some of the larger æstuaries were filled first with a deposit of fine clay of glacial origin, which seldom contains any organic material. In the Quinnipiac marshes this clay has been bored to a depth of over fifty feet without penetrating through it. The marine deposit overlaying this clay is only six or eight feet thick, but some parts are very rich in diatoms. Ancient marine deposits similar to those of parts of New Jersey, Md., Va., and California cannot be expected to be found north of the limit of glacial flow ; all such deposits were thoroughly ground up and carried away during the ice period ; but the time since then has been sufficiently long to produce an abundance of fresh water deposits,

and to line many of the shores with marine material.

Of the five deposits of fresh water diatoms which I have myself discovered in Bristol, Conn., three are from five to eight feet below the surface, and are each about two feet thick and cover several acres; the fourth is more narrow, about two rods wide, appears to connect two adjoining swamps, and the fifth is bisected by a railroad cutting. They lie at different elevations, from 400 to about 800 feet above sea level, and they all vary considerably in the contained species as well as in richness, though all are rich. They all show the characteristic grey ash color when dried and are light in weight.

The deposit which underlies the neighboring city of New Britian is not so rich, and contains more clay and sand; it is much more difficult to clean. It was discovered in digging for foundations for buildings, and varies in appearance, in richness and to some extent in contained species in the different localities where it has been dug up and investigated. This material requires examination under the microscope to determine that it contains diatoms, while the dried Bristol material shows at a glance to an experienced eye that it is diatomaceous. The Quinnipiac deposit also shows its diatomaceous character clearly when dry, but many of the shore deposits require an actual test to determine their value. It is probable that scarcely one in a thousand of existing deposits has ever been discovered or investigated, and collectors are in no danger of exhausting the supply of new material for many years to come.

Chemical Analyses of the So-called Infusorial Earths.

By ARTHUR M. EDWARDS, M. D.,

NEWARK, N. J.

It is interesting to compare the chemical analyses of the so-called "Infusorial Earths" which contain the shells

of Diatomaceæ, but I find records of only the following.

Hofman gives the analysis of the earth from Bohemia in the Journal fur Practische Chemie, 1864, thus :

	(1)	(2)	(3)	(4)
Ammonia	.03	.01	.34	
Potash	.02	.30	.24	.40
Soda	.30	trace	trace	
Magnesia	......	.43	.36	.05
Lime	.41	.44	.64	trace
Alumina, Ferric Oxide	.81	5.40	5.60	.91
Sulphuric acid	.12	trace	.54	......
Phosphoric acid	.24	trace	trace	.19
Organic matter	4.20	1.30	13.20	15.45
Water	13.30	10.90	7.00	6.00
Silica	74.20	80.30	72.60	77.00
	99.63	99.08	100.52	100.00

It is at Kutschind, near Bilin, Meistersdorp, Luisa spring in Franzenbad. No. 1 is from the upper layer Bilin ; No. 2 is from the lower layer Bilin ; No. 3 is from the Meistersdorf, and No. 4 is from Franzenbad.

These are all from the iceberg period, geologically. No. 1 of the "tripoli," as it is called, has a density of 1,862 ; No. 2, of 1,944. That is to say, it is nearly twice as heavy as water.

In the "Kiesulguhr zu Franzenbad," Ehrenberg discovered in 1838, what he called Cocconeis ? clypeus. It was afterwards, in 1841, called Campylodiscus clypeus, and was also found at Santa Fiore, Italy. This also belongs to the iceberg period. He also found, in the Franzenbad deposit, Navicula bohemica and N. sculpta. These are both varieties of N. amphisbæna of Bory de St. Vincent, 1826. Ehrenberg calls it N. ventricosa, 1830, and Kutzing calls it Frustulia depressa. It is not a Navicula, but a Neidium of Pfitzer.

W. Habirshaw analyzed the electrosilicon of Virginia City, Nev., and found it to contain :

Silica	81.08
Water, volatile at a red heat	18.44
Loss	.48
	100.00

This belongs to the Eocene period before the Oligocene Tertiary.

The California Infusorial stratum which was discovered by Bailey at San Pablo, near San Francisco, and near by which I lived nearly two years, and the Monterey and Santa Monica, is Oligocene though ranked as Miocene by most geologists.

Cooke gives an analysis of the Drakesville, N. J., Infusorial stratum, which is as follows:

Silica	80 66
Alumina	3.84
Lime	.58
Loss on ignition (organic matter and water)	14.01
	99.09

It belongs to the iceberg period.

In Mexico, near Socorra, within 12 miles of the city is a vast layer of Infusorial earth which is Eocene also. It seems to belong to the Occidental Sea, but it has not been investigated as yet.

Now we see that the Eocene does not contain any alumina and is not a clay, strictly speaking. But the iceberg period strata, both in Bohemia and in New Jersey are clays made by the breaking down of the granite. This is a difference between those from the Eocene and iceberg strata, and it has been rendered solid by the solution of the silica and not by the clay.

I do not know of them except the Franzenbad, which has been analyzed, and which is also known as Soos. In contains too large a quanity of Campylodiscus. In the Eocene of Utah lake and of Mono lake and of Pyramid lake, there is present a Surirella utahensis of Ehrenberg. But this is Surirella striatula of Turpin, 1828. It is the Navicula (?) striatula of Ehrenberg, 1838, but is a true Surirella. Surirella striatula is brackish and marine. It is also fresh water in one "Keiselguhr von Franzenbad." In fact, it sometimes occurs circular instead of elongated, and looks as if it were passing into

Campylodicus. Formerly Campylodicus clypeus was known as a fossil form and occurs in fresh water only. But it is a brackish form in the North sea and there recent. It is rare in the raised coast period of Newark, N. J.

Dall (1892, Bulletin, U.S. Geol. Survey, No. 34, p. 117) gives an analysis, by Prof. F. W. Clarke, of Bailey's Infusorial stratum at Ballast Point, Hillsboro County, Fla., as follows:

Silica	70.78
Alumina and iron	11.33
Lime	2.18
Water and loss	15.71
	100.00

Though he says: "An examination of this marl by Mr. Lewis Woolman, the well-known student of microscopic organisms, did not reveal a single diatom," Mr. Woolman sent me specimens of the marl in which I failed to observe diatoms also. Since then, I got some from Manitee, Florida, which is south about 35 miles from Tampa, Florida, and for which I am indebted to Mr. K. M. Cunningham of Mobile, Ala., and to Dr. D. B. Ward of Poughkeepsie, N. Y., which showed Diatomaceæ undoubtly in fossil condition. The fossil Diatomaceæ of a marine condition belonging to the Miocene or Oligocene period have not been detected on the Atlantic side of Fla., in Ga., S. C., N. C. nor in Ala., La., or Tex., nor in Mexico, but we may suppose it possible. That they will be found there, I am sure. I am only waiting for an opportunity to visit those localities myself, for now I am convinced that the fossil Diatomaceæ can only be found by what is called a Diatomaniac, in which category, I am proud to be known. The fossil Diatomaceæ of Newark, N. J., were found by myself although travelled over hundreds of times by dozens of students and I only found them when my attention was especially fixed upon them and when I had time to look for them.

In the Bulletin of the department of Geology of the University of California, 1893, is an article by Prof. Andrew C. Lawson on the geology of Carmelo Bay in California, wherein he speaks of the Monterey series, meaning the light colored rocks which contain the Diatomaceæ and which I have spoken of as found on the Pacific coast of California. He gives the results of a chemical analysis as follows :

Silicic oxide	86.89
Aluminum oxide	2.32
Ferric oxide	1.28
Calcium oxide	0.43
Magnesium oxide	trace
Potassium oxide	1.26
Sodium oxide	2.32
Ignition	4.89
	99.39

But this immediate specimen contained no Diatomaceous remains, and it is interesting to remark that Prof. Lawson, although classifying the rocks as Miocene, says : "The infusorial beds at Monterey appear, therefore, to be exceptional and not representative of the series as a whole," and it is suggested that perhaps the microscopic examination of the Monterey shale will show , as the geological analysis shows, it to be of volcanic origin—showing that we do not understand the geology of these rocks.

To Mount Certain Salts.

By NO SIG.

PARIS.

NITRATE OF URANIUM.

This is a most difficult salt to mount after having obtained a favorable specimen of crystalization, and requires the greatest attention to the most minute details in order to succeed in producing a satisfactory, well finished slide. A solution should be made nearly saturated and filtered.

Take a clean slide and run a ring of India ink on the back of the slide, as a guide for fixing the cover glass quite central. Place a few drops of the solution from a clean pipette in the center of the other side of the glass and put it away to dry on a shelf that is not quite level, which will cause a part of the solution to drain away to the bottom and corner of the slide; if the air is very moist it will not readily crystalize, nor give a good result, so that perhaps many attempts will have to be made before you succeed in obtaining anything worthy of being mounted; but when it is obtained the result will be found to compensate for the trouble taken.

Do not attempt to hasten the crystalization by warming the slide as even a moderate heat breaks up the crystals and spoils them. When, after examination under the microscope with the polarizing apparatus and the selenite film, you find that you have obtained something worthy of being preserved for the cabinet, put the slide on the turn-table, slowly revolve and scrape off the excess with a knife, having the disk a little smaller than the cover-glass which you intend to use; get your cover-glass well cleaned, and to preserve the crystals without any diminution of their brilliancy, put a few drops of the purest petroleum oil on the crystals, and drop the cover on removing the excess of petroleum with a small brush, but be careful not to remove any from under the cover-glass. If you cannot get it pretty clean, use a little spirit or benzole on the brush, wiping the brush after every stroke on a piece of paper, and removing the benzole by dipping the brush in, and removing the excess by draining it against the edge of the cup that holds it. The petroleum will soon dry off, when the glass can be fixed by a touch of Miller's Caoutchouc Cement at each side, putting as little as possible on; you must see that the cover is exactly in the center of the ink circle before fastening down.

In about a quarter of an hour the cement will have hardened sufficiently to enable you to run a ring of rather thin Miller's Caoutchouc Cement around the glass circle without disturbing its position; it is well to see before ringing it that the cover is full of petroleum. Should any have been removed by the brush or by evaporation, take a clean steel pin, dip it in the petroleum and place it against the part of the cover-glass that requires filling, when it will run under without there being any fear of forming air bubbles, and none will spread on the glass slides.

After having tried to mount this preparation without success in every medium I thought of—pure balsam, balsam dissolved in benzole, chloroform, etc., thus, dammar and castor oil, I thought of trying petroleum oil and found that it had no action on the preparation. Slides that have been mounted for some months are as good as on the day they were finished.

It is well that I should mention here a turn-table that has been invented for some years, but does not seem to be well known. It is called the concentric turn-table and is made by Aylward of Manchester, England, it is composed of an outer ring which revolves on the inner ring, each has two brass pegs, which, on turning the ring, clip the slide whatever width it may be within certain limits, and must hold it absolutely central, so that, on replacing the slide to finish, it always returns to a central position. The slides must always, of course, be the standard length of three inches.

Scheme for Imbedding, Sectioning, Staining, and Mounting.

By W. S. Miller,
Madison, Wis.

Specimens may be fixed in Flemming's chromo-aceto-osmic acid, 10 per cent nitric acid or a saturated aqueous solution of

corrosive sublimate and the hardening completed in alcohol; or they may be placed at once in a large quantity of Muller's fluid and after two weeks transferred to alcohol. After the hardening is complete keep the specimens in 80 per cent alcohol. The size of the specimen should be small and the quantity of the fixing fluid large; this rule must be faithfully followed if good results are to be obtained.

1.—When Paraffin is Used.

80 per cent alcohol, (*a*).
Stain, (*b*).
Wash.
Absolute alcohol.
Clear, (*c*).
Paraffin.
Imbed.
Section.
Fix on slide, (*d*).
Xylol, (*e*).
Absolute alcohol.
Stain, (*b*).
Wash.
Absolute alcohol.
Xylol.
Balsam.

2.—When Celloidin is Used.

80 per cent alcohol
Absolute alcohol.
Alcohol and ether.
Dilute celloidin.
Saturated celloidin.
Imbed.
80 per cent alcohol, (*f*).
Section.
Stain, (*g*).
Wash (Then glycerine will terminate if used here).
95 per cent alcohol.
Eosin-alcohol (*g*).
Oil of origanum cretici; or oil of cloves (*h*).
Balsam.

a. If sections are to be stained on the slide after imbedding in paraffin, pass at once to absolute alcohol; if the specimen is to be stained *in toto*, pass to stain, etc.

b. For staining *in toto* use Grenacher's borax-carmine, alum carmine or Delafield's hæmatoxylin. For staining on the slide use either the above, or, preferably, some one of the aniline colors. Specimens hardened in chromic or osmic acid mixtures take carmine stains badly.

c. Clear in either cedar oil, beech-wood creosote or chloroform. Avoid clove oil, as it makes the paraffin granular.

d. If the specimen has been stained *in toto*, use Schallibaum's collodion-clove-oil fixative; if sections are to be stained on the slide, use Mayer's albumen fixative.

e. If the specimen was stained *in toto*, pass at once to balsam; but if sections are to be stained, pass to absolute alcohol, stain, etc.

f. After imbedding and previous to placing in 80 per cent alcohol, it is well to place the block for a short time 1-2 hours in chloroform. This prevents the formation of bubbles and makes the celloidin more uniform in consistency.

g. For routine work use Delafield's hæmatoxylin and eosin-alcohol; this gives a double stain. Any other stain may be used, but some anilines color the celloidin intensely. If other stain than hæmatoxylin is used, the eosin-alcohol may be omitted.

h. If it is desired to remove the celloidin from the section, clear the specimen in clove oil. With delicate sections it should not be used.

Is the Paraffin Imbedding Method Better Than the Celloidin?

By PHILIP JAISOHN, M. D.,

Demonstrator of Histology in Medical Department of Columbian University.

Before discussing the respective merits and demerits of each method, it is necessary to describe briefly the technique of both, in order to discuss them intelligently.

The paraffin method was first used by Fredericq, and described by Schwalbe in 1886. (Anatomischen Anzeiger). Subsequently, Wm. C. Krauss described more fully with some modifications, in 1888. (Fortschritz der Medicin, 1888, No. 16). The method is as follows:

Put the fresh tissue first in 75 per cent alcohol, and keep it there for 24 hours; transfer it to 95 per cent alcohol, and let it remain for 24 hours; then again in absolute alcohol for the same length of time. Remove the alcohol from the tissue by soaking it in pure chloroform for 24 hours; transfer it to melted paraffin mixed with equal quantity of chloroform, leave it in this for 24 hours; then change to pure melted paraffin. After 24 hours or 36 hours, imbed the tissue in a paper box or metalic mould with paraffin round it. It hardens in 15 or 20 minutes, and is ready to cut.

To stain the paraffin sections, the paraffin in the tissue must be dissolved by oil of turpentine and then remove the turpentine by dropping the sections in 95 per cent or absolute alcohol, then to water, and the stain. Absolute alcohol is used to dehydrate the sections and the clearing agents generally used are turpentine or oil of clove. Celloidin method was first introduced by Duval in 1879. (*Journal de l'anat. et de la physiol. T. XV.*) Subsequently employed and described by P. Schiefferdectes (*Archiv. f. anat. u. Physiol.* 1882). This method has but one point in common with the paraffin process: the tissue is infiltrated with the imbedding mass; the

paraffin sections are cut dry while the celloidin must be cut under, or flooded with weak alcohol. Harden and dehydrate the tissue in absolute alcohol, then keep it in thin celloidin solution for 24 hours, (15 grains of Shering's celloidin dissolved in 100 cc. of equal parts of absolute alcohol and ether). Then transfer to a thick solution which should be made containing enough celloidin to secure the consistence of a thick syrup. Let it remain in this solution from 24 to 48 hours.

Fasten the tissue, by the thick solution, to a cork or better, a small block of wood; (care being taken that a stratum of celloidin lies between the tissue and the cork or wood) and a number of layers of celloidin should be placed around the tissue. In a few minutes the celloidin will be hardened by the evaporation of alcohol and ether, and it is ready to cut.

As it has been mentioned above, the celloidin sections are cut under a stream or flooded with weak alcohol (50 to 70 per cent). These sections can now be stained and dehydrated in 95 per cent alcohol and cleared in xylol, oil of bergamot or cedar oil. In either method the tissue can be stained before dehydration and imbedding. In some cases staining in mass is preferable to staining in sections.

In reading over the two methods, one will notice the simplicity of the latter process. It is an excellent method, especially adapted for certain lines of work in the central nervous system and special senses, and possesses the advantages over paraffin of requiring less attention, and no heat for its successful manipulation. It gives sufficient support to the tissue, and its transparancy (*verre elastique nenut sie Duval*), does not interfere with the topography of the field.

But in using this method, the sections are thicker than those of the paraffin and it is not applicable for serial

sections; and there is more liability of tearing and wrinkling during manipulation.

The paraffin method requires more attention (1) complete dehydration; (2) thorough infiltration; (3) proper temperature for the paraffin bath, which should never exceed 50 C.

In handling loose sections, some delicate tissues will fall apart after removing the paraffin from them, but this can be remedied by application of a fixing mixture such as the gum arabic, (Florgel Schultze), Celloidin, oil of clove, (Schallibaum), and weak gelatin solution (Gray). The object is to replace the support afforded by the paraffin, by attaching the sections to the slide before removing the imbedding substance. By this means, we obtain a secure attachment of the sections to the slides in all solutions necessary for the various manipulations of staining and mounting, and complete flattening or expansion of the sections before their final attachment to the slide.

If the above points are carefully observed and followed, I think the paraffin method possesses many advantages over the celloidin. In this, the sections can be cut as thin as it is required, and by the fixing method the subsequent manipulation is easier, the sections can be handled almost with impunity, as there is no danger of tearing or wrinkling, especially when serial sections are to be cut, this is the only method which will answer the purpose.

Prizes.—At the 16th annual meeting of the Missouri State Pharmaceutical Association to be held at the Excelsior Springs, June 12–15, a prize will be given as follows:

A set of twelve Botanical Microscopical Mounts for the member of the Association identifying the largest number of them by the aid of the microscope. Presented by Dr. H. M. Whelpley, of St. Louis, Mo.

EDITORIAL.

Spoiling Lenses.—Unfortunately a fifty dollar lens may be spoiled in ten seconds. The very carefulness to wipe it is sufficient to spoil it if rubbing is indulged in with gritty paper or cloth. A lens must be cleaned but not by rubbing. Rubbing generates electricity and the electrified glass attracts dust. New velvet may be used but some soft skin like chamois is safest.

Heat and cold must be avoided for they cause expansion or contraction of the mounting and may produce displacement of the lenses.

Solar light acts upon all kinds of glass; it imparts a slight coloring that has been studied by Faraday, Bontemps, Fresnel and others. It is rarely noticed in optical glasses but when discovered, the lenses may be worthless in case their properties have undergone change.

Moisture must be guarded against for it readily condenses on the glass when present, being attracted by the large proportion of alkalies used in the manufacture of the glass to impart transparency.

Czapski's Theory of Optical Instruments.—A well informed German physicist, when asked if any book had been printed during the year of special value, having at first said no, upon second though said: Oh, yes.—Czapski's *Theoric der Optischen Instrumente*—(*Nach Abbe*). This was unintentionally a very high compliment and calls for some notice. The writer shows perfect familiarity with the writings of Herschel, Smith, Lloyd, Airy, Rayleigh, Dallinger, Pendlebury, etc. Practically he had the great advantage of practicing upon all his thories in the great works of Zeiss in Jena where he is scientific adviser and technical director. His intercourse with Prof. Abbe, with skilled workmen, and his access to the best machinery and materials, all conspired to educate him for this task. His article in Van Heurck's last book on "The Future of the Microscope" introduced him to us in a pleasing manner, and his visit to the Columbian Exposition served to make his name familiar to American microscopists.

His first chapter is on geometrical optics; the second and third and fourth on the geometrical theory of optical images, the

fundamental properties of lenses and systems of lenses, and the theory of spherical aberration. Chapter five is on the theory of achromatism and chromatic aberration. Chapter six is on prisms and system of prisms; seven is on stops and aperture and properties of an optical system, which depends upon aperture, such as penetrating power and brightness; nine is upon the principal types of optical instruments, and ten treats of the methods for determining the constants of optical instruments. Much of the data is new to science and a distinct advance in physical research. The account of Abbe's theory of microscopical vision is not published, but withheld for a separate volume.

The Beaver Creek Meteorite.—It fell May 26, 1893 about 4 o'clock a. m., at Beaver Creek, West Kootenai District, B. C. It is of gray color and granular structure. In thin sections under the microscrope Merrill states that it presents no features not common to such stones. Structures are recognized similar to the meteorites of Madras, Homestead, Dhurmsala and San Emigdio. He says: "There are the usual monosomatic and polysomatic chondri sometimes of olivine alone, enstatite alone, or olivine and enstatite together, in granular or porphyritic forms with glassy base or radiating and barred forms. The olivines not infrequently occur with interiors made up of small rounded granules imbedded in a glass base, but extinguishing simultaneous with the outer portion."

A chemical analysis will be found in the American Journal of Science for June.

MICROSCOPICAL APPARATUS.

King's Glycerine Jelly.—Dr. King sends us the following regarding his improved formula. Glycerine Jelly is the best mounting medium where shrinkage is to be avoided, protoplasm preserved, and the finest differentiation of details to be attained. The objections to its use arise more than anything else from a lack of patience to overcome some slight difficulties in manipulation.

This Glycerine Jelly is made with an expensive gelatin from the swimming bladder of the sturgeon, a very different thing

from the cheaper and softer gelatins in common use, manufactured from the trimmings of slaughter house pelts.

It is filtered. It is perfectly transparent, and will not change color in the slide. Unlike glycerine it stays where you put it.

DIRECTIONS.

1. Work in a warm room, the jelly is hard and cools rapidly.

2. Heat, and keep hot while using, in a warm bath.

3. Mount from chemically pure glycerine and 75 per cent alcohol, one volume of each, kept till perfectly homogeneous, and filtered.

4. Warm the slide and place the jelly on it with a glass rod, kept clean.

5. Place the object in the jelly, being sure that it is well covered. This may require an additional drop.

6. Hold the object in place and drain the slide to get rid of the glycerine and alcohol.

7. Cover the object again with jelly and examine carefully under the dissecting microscope for air bubbles, especially for *stowaways*. Air bubbles should be worked off with a dissecting needle and not picked out with the forceps.

8. Cover the object again—take the cover glass between the thumb and finger,—breath on it,—cover it well with jelly,—take it by the edge with the forceps,—turn it over quickly,—place it gently on the object at an angle and apply a clip.

If it is desirable to mount more than one object in a slide, place them in just jelly enough to cover them safely from air, and give time to harden, after which an additional layer may be added and the cover placed as before and held with a clip Next pass the slide over a spirit lamp till the entire mass of jelly is melted. If the right quantity has been used the objects will not slide out, or if they become displaced push them back again with a slip of pointed paper slightly moistened in the mouth. No number of directions will supercede patience, practice and good judgment.

A Modified Microscope.—Mr. Hodgson, of Birmingham, England, has had Swift & Son construct an instrument with the following modifications:

1. The drawtube and milled heads are of aluminum.

2. The substage is suspended on a pivot to swing out of the optical axis when not in use.

3. The tail piece swings forward, and the mirror being attached by a saddle and crank arm, can be adjusted for use above the stage.

Increasing Angular Aperture.—The owners of high angle objectives are indebted to Dr. Henry G. Piffard of New York, for valuable hints as to the use to which such lenses can be put, in which the angular aperture can be considerably increased.

He found that a 1/4 inch water immersion having an angular aperture of 143°, would also work perfectly well with cedar oil, and more recently he discovered that by closing the systems as far as the correction collar would permit, and shortening the tube length to about 155 mm. that the objective would correct perfectly with mono-bromide of naphthalin. By these means he increases the aperture to N. A. 1.56.

Dr. Piffard made several experiments with other lenses on this basis, and found that most of them but not all, would respond to this treatment. In order to obtain the benefit of the full aperture here described, it is necessary to have at command adequate substage illumination, and of course the objective must have the collar adjustment.

I have a dry 1-6 inch of 165°, and have made a few experiments with it, following Dr. Piffard's suggestion and find it can be used as a very good water immersion lens. Dr. Piffard concludes by making a suggestion as to what an ideal immersion lens should be for general bacteriological, biological and histological work. Thus, it should have the largest practical angular aperture and be corrected for either the 160 mm. or 216 mm. tube, and a cover-glass not exceeding .20 mm. The adjustment should be arranged to close a little beyond the mono-bromide point, and open a little beyond the oil point, so as to permit the objective to be used at will for either fluid.

JOHN MICHELS.

A Cheap Instrument.—T. S. Middleton has made a microscope for $5.45 which he says any one can duplicate.

Take a board two feet long, four inches wide, one inch thick. On its edges screw paralled strips having beveled edges. In this slot, the various parts slide during adjustment. The funnel is

an ordinary tin funnel deprived of its lower end and painted dead black inside. To its under side is soldered a strip of tin, this forming a foot. It is screwed upon a piece of wood which has been previously fitted in the above mentioned slot. Behind it is the slide holder, made of very light wood and secured to a base fitted in the slot. The upright has a hole cut through it about $\frac{3}{4}$ inch in diameter, through which the image and light pass. Upon the face of the upritgh are two spring fingers for holding the slides.

Behind this is the brass tube which cost 45 cents. It consists of a piece of thin brass pipe, $2\frac{1}{2}$ inches long by 2 inches in diameter. It has a society screw cut in one end so that it may receive any objective. The inside of the tube must be painted dead black.

A ring is worked out of a piece of wood of not less than one inch in thickness. The thickness prevents any binding. Cut a small notch for the rack which keeps the tube from turning.

There is a small pinion that operates the rack. It is therefore necessary to cut the ring in two at the bottom of the rack. This is to allow for the cutting of the recess for the pinion and the shaft for the thumb piece. This is also fastened to a base piece that is fitted into the slot.

The object glass is simply a ground glass. This takes the place of the ordinary eye-piece, so that instead of viewing an object with one eye through a $\frac{1}{8}$ inch aperture both eyes are used as in ordinary vision. The ground glass is 6 inches square. The image can be seen on either side of the ground glass. If any white material, as muslin or cardboard, be substituted for the ground glass; the image can be seen on only one side. If the ground glass and holder be removed and the instrument be pointed toward a sheet suspended from the wall; any image of any desired size may be projected thereupon as in lantern views.

The center of the funnel, the hole in the slide holder and the lens, which may be any low power, must all be in line.

Any light from a candle to a lime light may be used but a cheap magic lantern lamp with two wicks is very good. One can be made out of a tin cracker box by cutting a hole in the side on a level with the flame and by inserting a reflector opposite the hole.

The apparatus can be used in photography by removing the

ground glass and substituting a small camera devoid of lenses. Focus very carefully in this case. Use the Carbutt-Ortho plate, $3\frac{1}{4}$x4 inches and expose $1\frac{1}{2}$ to $2\frac{1}{2}$ minutes according to strength of light. Develope with Hypodeveloper.

MICROSCOPICAL MANIPULATION.

Snowflake Pictures.—Anthony's Bulletin says that a Mr Sigson has photographed snowflakes as follows:

A Zeiss microscope, fitted to a long camera, was placed at a considerable angle in the attic of a house, near to the window. To gather the flakes separately, a thick cloth was laid in that part of the window where but few flakes fell. After selecting a flake by the aid of the microscope, it was placed in a net made by gumming cotton thread across a hole cut in a card; this card being placed in position under the microscope. The lighting should be from the side, and should be arranged beforehand, so that half of the field is lighted. With a magnification of 15 times, an exposure of 2 to 5 seconds was found necessary. To prevent the snowflake being melted by the breath of the operator breathing must be carried on through a curved tube.

Aniline Stains may be removed from the skin by acid or hypochlorite, but the better mode is to wash first with a 5 per cent solution of sodium chloride, then with hydrogen peroxide and finally to rub with a cloth moistened with alcohol.

Ringing Mounts.—For ringing mounts there is nothing neater, cleaner or more convenient for use than liquid shellac. Use the liquid pure without any admixture whatever. While the slide is upon the turn-table apply the liquid with a fine brush and finish with the point of a knife blade. Rings thus finished look almost as if made of solid glass. If a fancy mount is desired, paint a design on the slide and run the ring over it, as above. These rings make a handsome finish, which every one will admire. One can prepare the liquid himself. It may be obtained at a varnish factory or procured from dealers in microscopic supplies.

Bleaching Animals and Sections Fixed with Osmic Mixture.—Dr. Carazzi, of the Spezia Civil Museum, gives the

following "oxygenated-water method" for getting rid of the blackening caused by osmic acid treatment:

The peroxide of hydrogen becomes reduced at contact with organic matter, and this is bleached by the oxygen. But the oxygenated water is very instable and in a short time the peroxide of hydrogen is converted into water. By the chlorine method, heat was necessary and it has an injurious effect upon the tissues.

The difficulties are obviated by the peroxide of sodium method. The compound is a yellowish powder; when put in water, oxygen is evolved and the liquid becomes alkaline. But if the water is mixed with acid, the liquid remains neutral, because the soda combines with the acid.

Large quantities of peroxide must not be put in a small quantity of water, nor mineral acids be used, otherwise a violent reaction is set up, oxygen being evolved in large amount and the molecules of powder being thrown all around.

The acids more convenient for use are tartaric and acetic. For bleaching, use the following: Put a solution of tartaric or acetic acid (10 per cent) in a vessel if for animals, and in a test tube, if for microscopical sections, sink a small quantity of the peroxide, add slowly 70 per cent alcohol at the surface of the water, put in the object. The oxygen escapes from the water, rises quickly to the level of separation of the two liquids and dissolves in the alcohol, slowly bleaching the specimen which is supported in the alcohol.

MEDICAL MICROSCOPY.

Leprosy.--In a microscopical section ôf a leprosy knob, two sorts of bodies may be seen: 1st, round cells, having in their chief mass the size of white blood corpuscles, clear and small grained, not always provided with a visible nucleus; sometimes somewhat aggrandized, gulf shaped, of the character of round cells, although sometimes spool-shaped and provided with processes. These irregularly shaped specimens show a stronger granulation. 2d, flat cells, partly in form of knolls, partly of stars, which are especially distinguished from the first kind by their pale clear nature and a clearly visible, oval, water-roll-like,

bisected and already doubled nucleus. It is thus possible to diagnose leprosy with the microscope.

A Typhoid Epidemic in Buffalo.--Dr. Cary reports the cause "a contamination of our water." The cases were spread quite evenly throughout the city, hence milk or food could not account for it. Turning to the city water supply the proof is clear. Much of the winter, water was distributed from the Bird Island Inlet. Just preceding the epidemic, the water in Niagara river was low and the Bird Island Inlet opened. The city Bacteriologist has examined the water and reports abundant contamination of the Niagara river at this inlet. He has found Eberth's typhoid bacillus there in abundance. Over 500 cases of typhoid occurred in the city in one month from this cause.

Were one per cent of the doctors organized by the city into a Health Board for the prevention of disease, a large part of the business of the rest could be destroyed.

Gonorrheal Infection.—It has been shown by Brewer that in a case of gonorrheal infection, six years had elapsed during which no secretion could be pressed from the urethra, yet the microscope showed several characteristic colonies of gonococci. Six weeks afterward, contrary to advice, this man married and in two weeks, communicated the infection to his wife in whom it went through the various stages of inflammation and resulted in abscess.

Toxins—are alkaloidal poisons produced by the vital processes of pathogenic bacteria as alcohol is the product of the growth of the yeast plant. When produced in the system by the bacteria their poisonous effects are the symptoms of the particular disease that the particular microbe produces. Produced in the laboratory the toxin of lock jaw has been demonstrated to be fatal to mice in a dose of .0000005 gram. If equally poisonous to man, a fatal dose would be one two-hundred-and-eightieth grain. From this one can judge what fearful weapons the bacteria possess in their specific poisons. (Beiger and Cohn, in *Zeitschrift fur Hygiene.*)

Health Conference.—At the conference of Health Officers of Michigan, June 14 and 15, tuberculosis was the prominent theme. Dr. V. C. Vaughan gave demonstrations of Tubercle

Bacilli, methods of growing and staining them and methods of inoculating animals with the bacilli as well as of examining animals dead of the disease.

BACTERIOLOGY.

Tuberculosis Culture Medium.—Used in the Department of Agriculture: Distilled water, 1000 c.c., (1 quart); magnesium sulphate, 0.2 gramme (3 grains); acid potasium sulphate 1.0 gramme (15½ grains); sodium chloride, 10 grammes (2½ drachms); asparagin, 2 grammes, (31 grains); glycerine 70 c.c. (2¼ oz.). E. A. DE SCHWEINITZ.

BIOLOGICAL NOTES.

Living Nerve-Cell During Stimulation.—Hodge has made a microscopical study of the nerve-cell during the process of fatigue. He finds that the nucleus shrinks, when stimulated under these conditions, more rapidly than when the ganglion remains in the animal's body. Decrease in size is at first rapid, then slower and more rapid again. The slowing up at no time amounts to stand-still or to partial recovery. The controls shrink very little, 3 to 8 per cent as compared with 60 to 73 per cent in the nuclei of stimulated ganglia. Granules and oil-droplets have been seen to disappear from the cell protoplasm during stimulation.

DIATOMS.

Washing Diatoms.—To avoid the use of acid in clearing diatoms a correspondent of a country publication says, put it in a jar of water and shake it for *an hour*. Evidently the time of this man is of little value and the space of the editor equally so.

NECROLOGY.

Dr. N. L. Mallory of Rochester, N. Y., died April 28, 1894, from an over dose of chloral. Age 46. He had devoted much time to microscopical and bacteriological investigations, was a member of the American and Royal Microscopical Societies. In 1892 and 1893 he was a member of the Rochester Board of Health, and as such worked hard to eradicate tuberculosis.

Dr. Corydon L. Ford of Brooklyn, N. Y., died April 14, 1894, being over 80 years old. When he was connected with the Geneva Medical School, many years ago, a microscope was imported from Paris; and, when it arrived there was not a professor, nor any one else except him, who could put it together and use it. Now every first course student is obliged to learn its use. He was one of the best teachers of anatomy this country has known.

MICROSCOPICAL SOCIETIES.

Ottumwa Microscopical Society.

The last three meetings of this Society have been taken up with the microscopical examination of urine. Its physiological and pathological characteristics have been studied. Some very fine specimen have been shown containing casts of various kinds of crystals, cells of blood, pus, epithelium, etc., the study having been with a view to diagnosis. Three meetings previous to the last three were given to the study of hair, specimens of the hair of a great many animals and human hair were made. Cross and longitudinal sections were made and studied. Much attention was paid to the differentiation of human hair from that of animals because of its interest in medical jurisprudence. The collection made by the society is interesting. Some of the specimens are beautiful.

Quecket Microscopical Club.

March 16, 1894.—Mr. Morland presented 37 slides of Diatoms. C. F. Rousselet presented a series of pond-life slides. Edinger's drawing and photographic apparatus was shown by C. L. Curties. Messrs. Swift & Son exhibited the new biological microscope. Mr. Nelson had a paper on the Determination of the Foci of Microscopical Objectives, Lantern and Camera lenses by Arithmetical Formulæ. H. W. King's paper was on Amœba. An excursion was announced to the Royal Botanical Garden's in Regent's Park.

Birmingham Natural History and Philosophical Society.

March 6, 1894.—In the Microscopical Section Mr. Wilkinson exhibited beetles found in the rotten wood of a large ash tree

also wood destroyed by them. Mr. Udall exhibited scorpions from Egypt. Mr. Hodgson showed a new microscope, and a type slide of sponges prepared by Sinel. It showed Clathria seriata, Halichondria panicea, Tethya lyncurium, Pachymatisma johnstonia.

J. E. Wright exhibited Zeiss's apochromatic objectives and commended them as superior to all others.

Sheffield (England) Microscopical Society.

May 1, 1894.—Professor Denny gave a demonstration of the "development of a bird." The structure and composition of the egg were explained; then, the development of the chick embryo during incubation. A dozen slides of embryos of different ages were shown, these ranging from a few hours to five days, the "branchial clefts" being well marked in some.

The following process of preparation of the slides was novel. The embryo is washed in clean water twice, then placed in osmic acid (1 per cent solution) for a short time. This fixes and also stains a brownish-black; then soak in Muller's fluid for 24 to 48 hours. Then clear in clove oil. Mount in balsam. The differentiation in structure caused by the osmic acid is complete, being far superior to hæmatoxylin or any other stain.

NEW PUBLICATIONS.

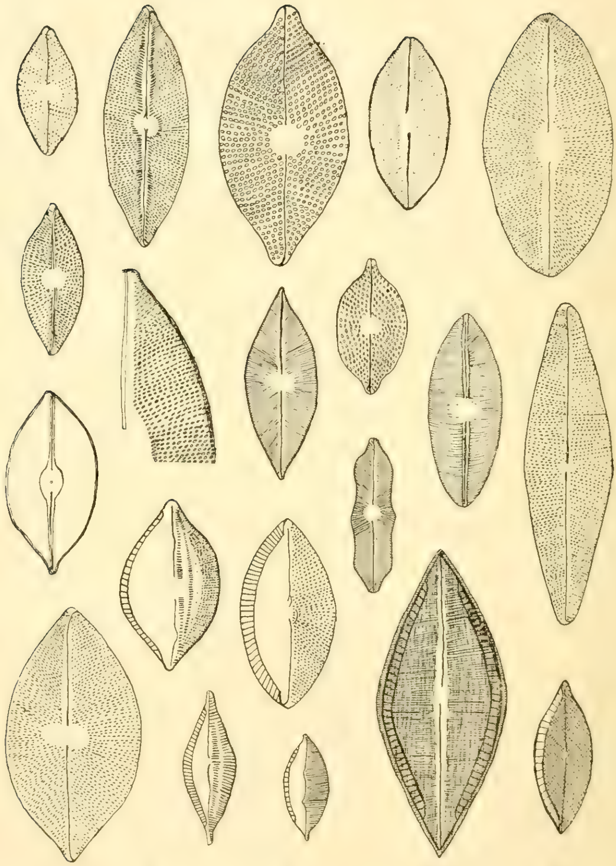
The Psychic Life of Micro-Organisms.—By Alfred Binet, pp. 125, 12 mo., 25 cents. Chicago. Open Court Publishing Co.

The author holds that all higher animals are merely aggregations of colonies of micro-organisms, each of which is endowed with psychical attributes. He explains how these organisms sustain themselves and seize food, resist poison, construct cells and maintain life.

Leitneria Floridana.—By William Trelease, St. Louis. Mo. Missouri Botanical Gardens. pp. 26, plates 15.

This is the description of a new tree found in Missouri by B. F. Bush in Nov. 1892. The plates are very fine and give illustrations of microscopic sections which we shall be glad to reproduce if Dr. Trelease will kindly loan the engravings.

It is one of the lightest woods known. Its structure is loose, tissues soft, heart wood entirely absent.



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SOME NEW AND LITTLE KNOWN DIATOMS.

THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. JULY, 1894. No. 7.

Studies in the Biology of the Diatoms.

BY K. M. CUNNINGHAM,

MOBILE, ALA.

Within the past year, various matters appertaining to the Diatomaceæ, drew my attention strongly to the question of their plant or animal nature; and in order to determine, or substantiate by personal observation, and experiences, I adopted as my initial task, the verification of the presence or absence of an investing protoplasmic covering or sheath; somewhat after the method suggested by Cornelius Onderdonk, as given in his article in "*The Microscope*" for August, 1890; but, after the inception of my experiments, I very soon ascertained that his method, while novel and interesting, *per se*, is not at all essential for the purpose in view, as I found a more direct and satisfactory method, and one which does not require the use of staining agents, or the effects of staining agents to determine the question.

After having made a number of satisfactory observations on the motion and other aspects of the living diatoms from special brackish and fresh water gatherings; I condensed my experiences in a paper which was read before the New York Microscopical Society and afterwards published in the Journal of the society in its Oct., 1893, issue; wherein I outlined my reasons for presuming that the Diatomaceæ properly belonged to the character of animal life classed under the Protoza, rather than to the Vegetable Kingdom or plant life.

The appearance of my paper on the subject resulted in

the receipt of a communication from the esteemed Diatomist, Prof. Hamilton L. Smith, of Geneva, N. Y., in which he advised me that he had read my paper, and called my attention to the fact, that I appeared to be unaware of what he had already written upon the subjects of staining, motion and gelatinous envelopes, etc., of the Diatoms; which papers were published in the proceedings of the American Society of Microscopists (latterly known as the American Microscopical Society) for its two meetings in the years 1886 and 1887 and which papers in the "reprint" are entitled, "A contribution to the life history of the Diatomaceæ, parts 1 and 2," illustrated with elegant, colored plates. Prof. Smith kindly sent me these reprints, and after careful and repeated perusals of the two papers, I became fully acquainted with their contents; but I found nothing therein, that could change or alter the views advanced by myself, as a result of my own previous personal observations; but on the contrary, the leading phenomena, recorded and figured by Prof. Smith, merely re-inforced and confirmed the views suggested by my own studies, and strengthened the arguments in favor of the Protozoan characteristics of the Diatomaceæ, as opposed to their vegetable or plant-like characters. So, that in order to verify the phases and observed characters recorded by Prof. Smith, and being also aware of the special specific forms studied, as well as figured by himself; I proceeded to a further and continued study of the living diatoms within reach, and circumstances duly favored my latter efforts, as I derived living material from four new localities not previously studied, or commented upon in my previously published paper.

It is from this material that I am enabled to extend the range of biological phenomena covered in Prof. Smith's two papers, to wit—In material derived from a moss-like water growth, found lining the edges of a run-

ning branch at Whistler, Ala., distant five miles north of Mobile, from which I expressed a quantity of liquid and sediment, (a test showing diatoms) which I saved in a bottle and took to Mobile on April 25, 1894 ; and on the evening of the day the specimen was secured, I made a concentration, and on microscopic inspection I found associated in the material species of Navicula major, N. divergens, N. viridis, N. viridula, Surirella splendida, Nitzschia sigma, Eunotia major, etc., and many species of true infusoria, ciliated, flagellate and non flagellate, associated with them, and also occurring in profusion. Beginning the study with some fine specimens of Navicula divergens, I noted that progression through the water was made equally as well when the sutural side was presented to view, as when the raphe bearing side was presented, and that generally when two frustules before complete autofission were travelling, the sutural sides were shown in preference to the valvular aspect and in their incessant change of place and position, grains of sand were snapped up, twirled about or transported by the *ectoderm*, (epidermis) or outside envelope of the frustule, each and every new Navicula showing something of interest in its actions.

Substituting for study, a Navicula major, I endeavored to note and observe the behavior of the living bacterial forms and minuter monad-like infusoria when passing the central nodules of the diatom, which are seen edgewise when the sutural zone is in view, as it was at these points that a form of molecular activity had been noted, as well as figured by Prof. Smith in his study, which indicated that very close to, and opposite the two nodules, small balls of indigo particles were being constantly formed, and after a while dispersed at these points. Instead of construing these molecular movements as attributable to a probable operation of the vorticellate power of ciliæ, which power is readily recognized in

many ciliated infusorians he partly suggested that the phenomenon reminded him of the well-known hydraulic experiment of causing a cork ring or ball to be revolved by the contact of a jet of water, or in his own language "of the rotation of the little balls of indigo, which is the most curious part of these observations there can be no doubt, and we may well ask, how is it to be explained ? One can very readily fancy a minute jet of fluid, issuing from the interior of the frustule through the little opening, or dot, and thus causing this rotation ; it is not unlike the well-known hydraulic experiment, which I have frequently exhibited of a cork ring clinging to the side of a jet of water and rotating ; but if this be so where is the water entering the frustule to keep up the supply ?"

While, after sufficient as well as attentive watching, I did not observe any particular motion of granules, bacteria or other matter moving in the liquid in the vicinage of the nodular areas, I did frequently see minute particles, such as bacteria transported by the movement of the exoplasmic sheath to where the nodular depressions occur, and they were then lost to view. (Not having any indigo pigment convenient at the time, I could not make the test by that medium of the formation of the little balls, but trusted more particularly to the minute infusoria, bacteria and other particles already in the liquid).

But while seeking for these effects, I incidentally observed that active monad infusoria were very readily entrapped by the exoplasm of the diatoms ; as I watched several thus caught, for a reasonable time, and I noted that they struggled with an incessant pendulum-like vibration to free themselves, but without avail. It is readily seen that failing to release themselves, they would finally cease to struggle and would settle down on the diatom, where it would probably be absorbed or assimilated by the protoplasmic covering of the diatom. Professor

Smith's important and interesting observation of the formation of the balls of indigo particles, was an incidental outcome of the experiment designed to determine the presence or absence of the investing gelatinous *ectoderm* of the diatoms; and the results of the experiments with indigo water-color liquid, as recorded and illustrated in both papers, aid in confirming and corroborating some of the results previously made known by Dr. Ehrenberg in his experiments on the injesting by infusoria and diatoms, of pigmentary particles, added to the waters in which they were studied and cultivated. As a consequence of what Ehrenberg had noted through this method of research, he decided that the Bacillariaceæ or Diatomaceæ were many stomached and were therefore classed by him among the Polygastrica (Infusorians), and thus placed under animal organisms. After his day, a school of English observers gradually beat down this classification, and eventually succeeded in having their views take precedence over Ehrenberg's; it is thus, as an outcome of change of views, through a species of mental evolution, that in the published volumes of the Challenger Expedition, if one desires to investigate the Diatomaceæ encountered on this three years voyage, and taken from the surface down to the abyssal depths of the great oceans, he will find in the volumes devoted to botany, the fruits of that expedition as worked up by the Count Abbe Castracane.

In Castracane's paper entitled "Generalites relating to the Diatomaceæ," he is absolutely silent as to the *motility of the exoplasm* or *protoplasmic covering of any of the motile genera*, and while he notes and calls attention to the backward, forward, and hesitating movements of the motile diatoms; he does not analyse the motion deeply and suggests that their motion is probably due to the drawing in and exuding of water from opposite ends of the frustule. This was his view as published (1884), and

closely resembles that of Rev. Wm. Smith, 1856; (see Carpenter) and analagous to the hypothesis enunciated by Prof. H. L. Smith at a Meeting of the American Society of Microscopists, 1887.

For some reason or other, not fully evident or reasoned out, and as appears in recent works on the Protophytes, Protozoa and Animalcular Infusoria, the motile genera of the Diatomaceæ are held to be "simple unicellular plants," or "lowly plant organisms" and are now very generally accepted as belonging to the plant or vegetable kingdom, without any further controverting or challenging as to the grounds or basis of evidence upon which this position is assumed and held. The works of Louis Figuier entitled "The Vegetable World," and "The Ocean World" throw interesting light on the subject of the labors of Dr. Ehrenberg, and other micrographers in connection with the history of the development of the biology of the Infusoria and Diatomaceæ which appertains to the questions previously under discussion, even to the question as to what does or does not constitute a stomach in an Infusorian.

But without regard to how much or how little Ehrenberg may have done in this special line of investigation, every one interested in the life history of the Diatomaceæ will be permanently indebted to Prof. H. L. Smith for the beautiful colored plates and the descriptive record of his observations, views, etc., thereto made accessible, and within reach of all American students of the Diatomaceæ.

Reverting to my observations of living diatoms. I next made a persistent study of several large Navicula nobilis, both in the single and twin or double states and observed very closely the appearances of the endochrome, and other contents, contained within the siliceous frustules, when I repeatedly verified in N. nobilis and previously in N. viridis, and divirgens, that the globular bod-

ies commonly referred to as "oil globules," have an independent motile power of their own, and when they are relatively few and scattered, and when not densely packed, the amplitude of oscillation of a group of three or four of the globules is readily followed, and in a group of three of these globules of the same or of various sizes, one of them may glide between the two others, making a cross over, thus reversing its position with reference to the two others which makes the phenomenon of motion very evident and noticeable. In reference to this, it may be noted that Prof. Smith, in referring to these so called "oil globules" always put an interrogation mark after "oil," thus "oil ? globules," thus indicating some degree of doubt or uncertainty as to their real nature or function but as the reagent *ether* dissolves them, the reaction suggests that they are of an oleaginous principle. These so called "oil globules" were the only possible morphological condition that could have induced or lead Ehrenberg to regard them as stomachs, as there is nothing else evident that has a structural shape that can be specifically made out excepting the nucleus, an organ which I have, so far, not been able to discriminate, identify or detect, the zone-like bands or layers of endochrome being apparently homogeneous or structureless and blending into varying shades of green or brown and gradually vanishing in the body of the transparent endoplasm which is usually colorless towards the middle of the frustule on either side, away from the nodular depressions.

Ordinarily at the median nodules, an indistinctly granular appearance is noted in the endochrome and it is within this area, the nucleus is to be looked for when the conditions favor glimpsing it. This area intervening between the nodules, seems to act as a partition, dividing the endoplasm into halves and within each half may be seen irregularly scattered, the globular bodies of various sizes and irregularly distributed within the more

fluid endoplasm. The Diatom is said to contain chlorophyll or a substance called Diatomin, but when we are studying one of the largest of the Naviculæ, we find that the general color of the whole frustule whether seen under a power of x600 or x1.200 is some shade of green or brown and that the globules themselves contribute most of the green color, independent of the darker stratified layers along the frustular zones as well shown when the sutural faces are shown in the field.

Now by comparison (in the relatively short study made for the purposes of this paper), I have incidentally observed many species of Infusorians (Protozoa) filled with green globules, commonly regarded as food balls, which are scarcely distinguishable from the greenish globules associated with the contents of the Diatomaceæ and as the Infusorians whirl, drag or surge along, the green globules tumble or roll about promiscuously or flow in processional streams, or remain in the same place relative to some adjacent organ, and sometimes the most minute Infusorians appear of a brighter green than diatoms ever appear. In an active Navicula major in the double frustular condition, and moving with its sutural sides uppermost, I observed that a brownish and rather dense layer of endochrome rested longitudinally upon and compressed three large diaphanous, colorless globules, between the layer of endochrome and the valvular face of the Diatom. The resistance of these three globules to compression gave the zone of endochrome a wave-like undulation, very striking in its effect, and which had not been previously seen in any previous studies of Naviculæ. This special case of the internal contents suggests a more complex morphological condition than seems to have been noted heretofore as appertaining to the general contents of the frustule.

Reverting to appearances observed by Prof. H. L. Smith in relation to the biology of the Diatom, the ma-

terial I had under study provided me with living specimens of Surirella splendida. A statement of some of the phases observed by myself will enable me to substantiate and verify a part of the interesting researches recorded by Prof. Smith. While searching through the heavier sediment, I found one large living specimen of Surirella splendida, the same form as figured and observed by Prof. Smith. I made a continuous observation of this form for more than one hour, and therefore got a good idea of its biological structure. While Prof. Smith and Dr. Carpenter both refer to the genus Surirella, or to particular species of Surirella, in terms such as to indicate that their movements or travelling power is insignificant in its character; I, on the contrary, found that they were fully as active as Navicula major, and required constant attention to keep them in the field of view under a one-sixth objective, and as I studied four distinct specimens of the larger Surirella on three different evenings, I found that all moved about the same as the larger Naviculæ and when they were not intercepted by debris of the slide, would readily move out of the field of view, the same as the other travellers do. Carpenter alludes to their movement, as a "lanquid roll" and Prof. Smith uses the terms "lazy sort of roll" and elsewhere refers to its "lanquid roll," but from my personal experience, I would assert that their actual change of place is as full of interest as that of Naviculæ and is of the same character, but with a greater tendency to lose its balance while in motion, thus suggesting the roll by a change of aspect in the position of the alæ.

While studying the motions of two of the Surirellæ, while in the field, on two successive evenings, I closely observed their behavior while working their way through masses of debris intercepted in their movement. The minute vegetal debris, etc., was simultaneously passed from their anterior prows backwards coutinuously along

both alæ and without apparent intermission until it took a retrograde movement, and backed away to a clear water space. While observing this behavior in one of the frustules, I also noticed that at one moment the Surirella turned upwards in the field, axially, to the tube of the microscope; and for the first time, I was enabled to understand the relation of the alæ to the sides of the frustule. If we conceive diagonals cut through a square, and projecting some little distance beyond the corners, we will then have a correct idea of their position with reference to the frustule, or in other words they flare out at an angle of about forty-five degrees to the sutural edges, or to the lined valves. I also noted clearly that in all four frustules, as examined, numerous spherical corpuscles were in active and constant motion. The first idea suggested by these appearances was that the spherical corpuscles moving within the endoplasm were minute monad-like Infusoria ingested by the Diatoms in their feeding, yet still retaining their motile power. Although I was quite familiar with Prof. Smith's references to these corpuscles, his paper did not prepare me for this surprise and I trusted to a careful observation for my own account, so that I watched them attentively in order to determine the apparent law governing their motions.

Also, bearing in mind the conjecture of Cornelius Onderdonk, that the life movements of the diatoms might be caused by some impulse on the inside walls of the cell, somewhat after the manner of the cyclosis seen in the cell structure of various well known water plants and Desmids; I very soon saw that this hypothesis had no value in the case of the Surirella studied by myself, as I found that the motile spherules went indifferently in any direction in the field of movement to which they were limited, two or more might pass each other going in opposite directions, and that they constantly crossed over

from the larger groups on opposite sides of the frustule, there being a rather wide clear space in which the spherules did not congregate but merely crossed and recrossed in their movements.

In Prof. Smith's figures of Surirella, these spherules are indicated rather sparsely, but in the first two specimens observed by myself they were quite numerous and of a uniform size. Outside of the actively motile spherules, there was nothing else of special interest to hold the attention, as the endochrome was apparently structureless, but showing different phases of opacity and density. Faint flecks or cloudlets of endochrome could be seen along the central portion of the frustules, a little more densely aggregated at its middle portion where the nucleus or nuclei are thought to exist, and which, as a partition confines the spherules, and apparently deters them from passing freely from one end of the frustule to the other end.

It happens with Surirella, as with most of the larger Naviculæ, that while they are being observed on the sutural face, they turn over, presenting the valvular or lined faces to the observer, when it is not then so easy to study the aspects of the motile spherules above referred to. Motile spherules of a similar character or behavior, were never seen by me in any previous study of any of the larger Naviculæ, and as Prof. Smith's specimens of Surirella, and those watched by myself, agree in all essential particulars, we may regard the spherules as a persistent character of the Surirella, although a period of at least twenty-five years has passed between the observations made by Prof. Smith and my own.

Prof. Smith, in following throughout the full process of conjugation of two individuals of Surirella elegans, followed the motile spherules, from the beginning of the point of fusion of the two separate endochromes and en-

doplasmic contents into a single sporangial mass, thus attesting a living transmigration of the motile spherules, and their re-appearance again on the completion of the mature sporangial frustule resulting from this conjugation; through which means, and by persistent observation, he was enabled to establish the fact that Surirella splendida, with its double nucleus, is the sporangial descendant of Surirella elegans, through conjugation alone.

From the above we may recognize the following phenomena, that as a result of conjugation, there follows a union of two pre-existent masses of living protoplasm, endochrome and contained motile spherules, and the rejecting or shedding of the inert siliceous frustular cases of the progenitors. These interesting facts suggest the following query. Can a single organism, arising from the fusing together of two distinct living units and presenting in its internal structure two distinct nuclei, be regarded as a unicellular or single-celled organism: there having been in the transition between the two states, no cessation, suspension or stoppage of vitality in the change from the dual lives to the resultant frustule?

Prof. Smith in his published papers, gives with full detail the phenomena occurring through the full period of conjugation, from its reception, until the sporangial frustule is fully formed, in which case we see that it begins its active life with the motile spherules indicated in their previous condition as filling the endoplasm of the diatom.

In reflecting over the nature of the movements made by the motile spherules, I felt that they must be actuated by a flagellum after the manner of the spermatozooids but not in the manner characteristic of the zoospores of Vaucheria and other plant algæ, and sometimes designated as coniocytes. They are top-shaped, flagellate bodies, which are known to richochet or spin around in a free state when ripe and freed from their parent cells.

To better illustrate the motion of the motile spherules of the Surirella, I could not easily recall any similar or analagous movements, except that seen when one observes colonies of tadpoles, in clear water branches, where the tadpoles may be seen in shallow places in crowded groups. At such a time, it will be noted, that there is a continual darting about, or mutual crossing to opposite groups, across an intervening space ; but, in the case of the motile spherules, the source of the propelling power evades the lens. I suspect that if the spherules could be examined after chance extrusion from the frustules, a flagellum, as in the case of Monas termo, might possibly be demonstrated with a suitable power. The reader will thus perceive that a variety of vital properties, and varied morphological phenomena are exhibited by a Surirella, during the course of its life cycle. This preponderance of intelligent and consecutive phases of protoplasmic and molecular activity encourages me to believe that the construing of all these varied vital phenomena, of such a simple nature as to lower them to the simplest forms of plant life, is done in contravention to their highly complex organization, as compared to many of the Infusorians classed under the Protozoa.

In the material secured for the use of the studies embraced in this paper, I had occasion to observe many kinds of Infusorians, Desmids and Rhizopods, and as the living Diatoms were more particularly my object of study I was enabled to study species of the following genera, viz; Surirella, Navicula, Nitzschia, Eunotia, Bacillaria, Cyclotella, Synedra ; and in six of these genera, I ascertained the absolute indication of the contractile and retractile power of the investing ectoderm or mantle of exoplasm, without the aid of staining agents or pigments, but simply by watching their travelling motions through the water on the slide. This method alone is sufficient for all purposes of demonstration of

the presence of the phenomenon which I have described.

As illustrating further some of the appearances, I observed Navicula viridis travelling in a group of four united frustules, convey simultaneously, three grains of sand on three of the frustules, one apiece, in the same direction, the three grains moving all in a line, and one grain of sand moving in an opposite direction all at the same time, three of the grains of sand being on top of the frustules and the fourth, under the frustule of the frustule that carried no grain on its upper surface. I also observed a large active Nitzschia sigma, constantly carrying grains of sand along both opposite margins, indifferently, and the selfsame Nitzschia caught up and conveyed along one of its marginal edges, two grains of sand which starting from the opposite ends met at the middle portion of the Diatom, the frustule while thus engaged, presenting its sigmoid outline.

I also witnessed living Bacillaria paradoxa, having two minute frustules of Gomphonema, attached by stipes to their frustules, and projecting out at right angles in a parasite-like manner, which rode backward and forth with every retraction or extension of the line of frustules, and whenever the Bacillariæ forced their way through matted debris, the minute Gomphonemas were bent back against the sides of the frustules bearing them, until the debris was passed, when they would stand out as before, thus proving that the stipes were firmly attached to the siliceous frustules bearing them, and that the stipes were flexible and acted like springs, and therefore limited strictly in their movements to the movement of their host.

I added lead pencil (graphite) parings to the water on the slide, and observed many small Naviculæ gathering up and rapidly circulating upon their frustules the particles of plumbago, constantly exchanging one particle for a new particle (this I did in lieu of any indigo). At

another time, I prepared a slide covered with a coating of very fine black pepper dust, with the object of noting the behavior of the living Naviculæ while in contact with the pepper. I observed that all of the diatoms previously alive, were inactive and apparently killed by the acrid or pungent principle of the pepper. From what has preceded, it is readily observed, that a large portion of the activity of the diatoms, while travelling on the slide, in the field of view, is passed in a ceaseless labor of moving around, catching up, whirling and dropping sand grains, and other minute particles.

Returning once more to the so-called "oil globules" in the various species of Naviculæ, etc.; while I stated that I had made a close study of their motile phases, I did not attempt to follow their morphological changes further than that; but from Prof. Smith's researches, I noted that he had followed species of Naviculæ through a complete life cycle, and that he observed that the motile globular masses coalesced and united together in the frustule, where they finally appeared as two large rounded masses, which were termed "the fully differentiated endoplasm," which give origin to the sporangial frustules derived from the conjugation of two contiguous frustules, or in like manner, of a single frustule performing the same function within the limits of its own frustule.

In my studies, as recorded herein, of the large Naviculæ, Surirellæ, and Nitzschiæ, I made use of a magnification of 300, 600 and 1,200 diameters, which latter case gave me by comparative estimate, floating images of the diatoms nine and twelve inches in length, and, in the case of Nitzschia scalaris, eighteen inches; but the general study was made with a power of 600 diametes, agreeing approximately with the dimensions of the species found in Schmidt's, Atlas and Wolle's Diatomaceæ of North America.

Microscopical Technique Applied To Histology.—IV.

[FROM THE FRENCH OF RENE BONEVAL.]

Continued from Vol. XIV, page 255.

FIXED CELLS.—To see the protoplasm, the lateral wings, and the nuclei, extend a filiform tendon on a piece of glass and fasten the two ends by paraffin. On the middle of the tendon put a drop of picro-carmine, and leave it for an hour in a moist chamber. [A saucer with a layer of wet blotting paper and covered by a bell glass makes a good moist chamber.] Wash and mount in glycerine. Apply gentle presure with the needle.... The anastomoses, which show the relations of the tendinous cells, are not seen in this preparation. For these remove the skin and place a portion of the tail in a large quantity of an aqueous solution of picric acid. When decalcified, harden by alcohol and gum. The sections, freed from the acid by prolonged soaking in water, are strongly colored by picro-carmine, and mounted in formic acid glycerine (1 per cent solution,) which should be allowed to penetrate the tissue very slowly. The tendinous fibres are de-colored, the cells and their prolongations are stained red....

APONEUROTIC MEMBRANES.

It is best to select a simple aponeurosis, that of a frog's thigh being elegant and most effective. Ranvier's method is the preferable one. Skin the frog and with the scalpel circumscribe a portion of the aponeurosis above the triceps muscle and take it off by the forceps. Remove, by brushing, the endothelial cells of the subcutaneous lymphatic sac and the pieces of muscle fibres adherent to the under surface. Carefully spread on a slide, partly dry it, stain with picro-carmine, wash till the yellow color has disappeared, and cover by thin glass supported on two sides by paraffin. Allow a drop of

acetic acid to run under, and, when the tissue is transparent, add glycerine. . . . .

SEROUS MEMBRANES.

Serous membranes present on the surface an endothelial lining readily demonstrated by silver nitrate. It is well to study the forms and relations of the cells in the perforated and unperforated serous tissues. For the former select the mesentery of the frog or of a young rat; for the latter the omentum of man or an adult rat. . . . Cut the omentum rapidly without touching it with the fingers. It is better, if not soiled by blood, to place it at once in the silver solution; otherwise wash quickly in water. . . Use a weak silver solution and agitate the membrane continuously while exposed to the sun. Wash in a large quantity of water, partly dry as described, stain with alum carmine, mount in balsam.

The connective stroma of these tissues may be studied thus:

1.—A piece of mesentery or of omentum is stained with picro-carmine, washed to remove the yellow color, and mounted in glycerine or in balsam. This will show the cells and the connective fibres of the mesentery; other parts we reserve for future consideration. In these preparations the cement between the connective fibres may be shown, if Ranvier's method be used. The membrane spread on glass is deeply stained with picro-carmine, treated with absolute alcohol, cleared by clove oil; make an incision with an exceedingly sharp razor, and upon the edge of the cut, between the fibres will be seen the pale rose-colored cementing substance.

2.—To demonstrate the layers of the mesentery described by Ranvier proceed thus. Introduce a slender pipette near a vessel in the mesentery spread on a glass plate. With the breath form a bladder; cut away the upper surface, the lower remaining adherent to the glass.

The latter is studied after picro-carmine staining. We thus obtain either the vascular layer or the non-vascular. In the latter the elastic network of the mesentery is observable.

3.—The pericardium, the pleura, the peritoneum, the synovial membranes are studied by transverse sections, made after alcoholic hardenings. Stain by picro-carmine; mount in glycerine with a little picro-carmine.

CARTILAGINOUS TISSUE.

The study of this is very simple. We will first study hyaline cartilage. A frog's xiphoid cartilage, the sclerotica, or the inferior angle of a salamander's or a triton's scapula may be used; in the last mentioned the cells are colossal.... Mount in a drop of aqueous humor.... Make sections from the fresh head of a frog's femur; in a drop of water can be seen the retraction of the protoplasm which gives each cell the stellate appearance which many authors have believed to be normal.

In fresh preparations the action of stains is easily studied. It is best to use sections of cartilage fixed by picric acid. A fragment is placed for 24 hours in a saturated aqueous solution of the acid; the sections are soaked in water until the yellow color has disappeared. Put some in alum carmine. After a few minutes, wash, mount in water, for which glycerine should be substituted by allowing it to run under very slowly. The nuclei are stained lilac red.

In this study Ranvier uses the purpurine extracted from madder. To prepare this, boil 200 grms. of water and 1 grm. of alum; when in ebulition add the madder and and continue to heat. In ten minutes filter while hot and add 60 c. c. of 36° alcohol. This should be used fresh as it will keep for only a few weeks. The sections are left in a few c. c. of the solution for 24 hours. Wash in water; mount in glycerine. The nuclei are stained red, the basis substance rose....

Osmic acid colors oil drops in the protoplasm black or deep brown. Sections may be put in a 1 per cent solution, or be exposed to osmic vapor after fixing by picric acid. Wash, mount in glycerine.

To stain the basic substance, guinolein blue is best. To a porcelain saucer of filtered water add a few drops of an alcoholic solution of the blue. The water at once becomes beautifully light blue with an iridescent surface, due to a precipitation of some of the coloring matter. In a certain time the water becomes decolored, the blue granules falling to the bottom. The water should not be too deeply colored. The sections, when entirely freed from picric acid, should remain there for a few hours. Wash carefully; mount in neutral glycerine. The cells are stained blue, the hyaline substance a beautiful violet. In preparations thus stained, and treated with the 40 per cent potash solution, we can obtain a blue color in the fat granules within the cell protoplasm.

Cartilage with branching cells from the head of a cephalopod may be studied in sections after fixing by picric acid. Stain with picro-carmine. . . .

Elastic cartilage should be studied in sections of the epiglottis of man or of the dog. A portion is fixed in picric acid for 24 hours, hardened in alcohol, embedded in gum; longitudinal sections are made and put into water. When freed from the yellow color, stain in picro-carmine, wash, mount in water, and allow to run under the cover a drop of the following, as suggested by Ranvier for the study of arteries:—glycerine, 50; saturated solution of picric acid, 50; formic acid, 1.

To study fibro-cartilage, section the semi-lunar cartilage of the knee; treat with picric acid, alcohol, gum. Stain with picro-carmine.

BONY TISSUE.

Two kinds of preparations should be made, one from

dry and macerated bones, the other from fresh decalcified bones.

SECTIONS OF DRY BONES.

Select a very white and dry bone, preferably an adult long bone. Avoid those with yellowish or translucent spots showing the presence of fat. . . .Fasten the bone in a vice and with a fine saw cut a series of plates as thin as possible, some longitudinal, some transverse. Rub them between two punice stones, taking care to turn the section occasionally and to keep it wet. When thin enough, wash it and smooth it on a whetstone. The best way is to hold the section by the finger tips and move it to and fro. By Canada balsam it may be fastened to a slide, or by gum to a cork, and so rubbed down, the cement being dissolved away when the bone is thin enough. . . .

TO EXAMINE THE OSTEOBLASTS FILLED WITH AIR.

Spread gum over both surfaces of a section and dry it rapidly over a flame. Heat a little old balsam on a slide and mount the section, cooling the balsam rapidly on a cold stone or marble. The gum prevents the entrance of the balsam into the corpuscles.

TO STAIN OSTEOBLASTS AND CANALICULI.—This is M. Ranveir's method and produces a true interstitial injection of the canaliculi and the oseous corpuscles. With a scalpel scrape a section made as described. It is then put in an alcoholic solution of aniline blue in a test-tube closed by a cork pierced by a hole in which is placed a tube a metre long. The solution is boiled on a water bath for 5 or 6 hours, the alcoholic vapors condensing and falling back to prevent the entire evaporation of the fluid. The section is smoothed on both surfaces on a whetstone wet with a solution of common salt. Mount in neutral glycerine.

To EXAMINE THE OSEOUS LAYERS.—It is possible to study these in the preceding preparations, yet we recommend another process. Dehydrate a section in absolute alcohol. Replace the latter by a drop of oil of bergamot, [clove, cajeput, or other essential oil] and when transparent mount in balsam. Here the osteoblasts and the canaliculi, filled with the resin, are hardly visible, but the arrangement of the lamellæ is very evident.

SECTIONS OF FRESH BONE.

A small fragment of the femur as fresh as possible is put in alcohol for a few hours, thence transferred to at least a litre of a saturated solution of picric acid, to which is added 2 per cent of nitric acid. Renew this liquid the next day. If the piece is very small (a condition indispensable to success), decalcification will be complete in from 3 to 4 hours. This will be when the bone is flexible and easily cut. Wash in a large quantity of water, which should be renewed until all acid is removed. Harden by alcohol; imbed in gum. Sections (made in different directions through the bone) are washed until the yellow color disappears, stained in picro-carmine, and mounted in carbolic acid water or in glycerine. This method is admirable for the study of ossification.

To study the part taken by the vessels in the formation of bone, inject the arteries of a kitten with the Prussian blue gelatine and put a femur for a week in 2 per cent ammonia bichromate. Decalcify in an aqueous solution of picric acid. Wash thoroughly, harden in alcohol, embed in gum, cut longitudinal sections and mount in balsam.

(*To be continued.*)

Cementing on Glass. To cement brass or any other metal upon glass, take one part caustic soda, 3 parts resin, 5 parts water, and rub up well with plaster of Paris equal in weight to half that of the above mixture.

Some New or Little Known Diatoms.

BY M. P.-T. CLEVE.

From Le Diatomiste.

SEE FRONTISPIECE.

[For the purpose of calling attention to the excellent work being done upon diatoms in Paris by our friend Tempere we abstract sufficient matter from M. Cleve's article to explain the plate and hope everyone who reads French will take that periodical. No diatomist can do without it.—EDITOR.]

1. Navicula gamma, n. sp. From fresh water in Guatemala, fossil.

2. Same, var. rectilineata. Brackish water at Camaroon, Africa (rare).

3. Navicula epsilon, n. sp. Marine. China and Japan.

4. Navicula alpha, n. sp. Marine. Japan.

5. Navicula eta, n. sp. Marine. Japan, Red Sea.

6. Navicula mœandrina, n. sp. Fresh water. Oregon. Fossil.

7. Navicula delawarensis. Brackish water Conn., mouth of the Delaware.

8. Enlarged view of same.

9. Navicula demerara. Fresh water. Demerara river, Surinam.

10. Navicula delta, n. sp. Marine. Ceylon.

11. Navicula tau, n. sp. Fresh water. Demerara river.

12. Navicula limicola, n. sp. Brackish water. Camaroon, Africa.

13. Navicula pi, n. sp. Marine. China.

14. Navicula grovei, n. sp. Marine. Oamaru, N. Z.

15. Mastogloia rimosa, n. sp. Marine. Bahama Islands.

16. Mastogloia obesa. Marine. Japan.

17. Mastogloia bahamensis, n. sp. Bahama Islands.

18. Mastogloia cuspidata, n. sp. Marine. Bahama Island.

19. Mastogloia antiqua, n. sp. Fossil. Karand, Hungary.

20. Mastogloia delicatula, n. sp. Bahama Islands.

All the above except numbers 3 and 7 are enlarged 800 diameters.

EDITORIAL.

Reviews of Microscopical Books.—A new book entitled "Practical Histological Botany," pp. 66, by Frederick Davis, B. Sc., of 26 Newington Causeway, London, S. E., and published by John Gower, 4 Lancing Road, Ealing, London, has just been announced in the Pharmaceutical Journal as follows:

"The only valid reason for noticing this book is to utterly condemn it and the system which encourages the multiplication of such mischievous publications."

Upon turning to the advertising columns of that Journal, we find Mr. Davis is an advertising patron and that John Gower is also; and yet the editor denounces their book in unsparing terms. Such a thing could hardly occur in America, (the more's the pity perhaps) but we cite it to illustrate the different customs prevailing in the two countries.

The American methods are shown by the following incident. A publisher recently sent to us a book for review. We inquired if he would like to begin an advertisement in the number in which the review would appear. He replied that he wished to see the review before he decided about the advertisement. We wrote the review and sent a copy to him. He did not like it and declined to advertise. He forwarded our slip to the author, who wrote to say that the review was quite unsatisfactory, two sentences seeming to reflect a little on his book. He also desired that much more of praise should have been added and a proof sent to him, for his approval before publication. In return for such favor he made to us certain offers.

Meanwhile, other notices of the book have appeared. The denunciations which it has received from a very prominent magazine are unmerciful and terrible in comparison with our faint criticisms. But a little country publication, which aspires to rival ours, has given the book unstinted praise. Consequences: the prominent magazine gets no new subscribers for telling the truth, and the author stands ready to aid the country ival in various ways.

In asking another publisher what he would do, we were advised to say anything the author desired, since it would bring positive advantage to us, and since telling the truth about the book would not secure us any appreciative support from our subscribers commensurate with the cost.

Subscribers and readers! What do you say? Do you approve the English or the American style and what will you do about it?

Authors and advertisers! Will you withhold advertising as a punishment for our telling the honest truth, as we see it, about your books? We want a lot of answers to these questions by first mail.

MICROSCOPICAL APPARATUS.

Instantaneous Photomicrographs.—Apparatus has been prepared by Chas. Baker, London, for making instantaneous photomicrographs and viewing the image until exposure is made. The apparatus consists of a case containing a metal shutter, carrying a prism, and connected with a pneumatic release. When this shutter is set, the image in the microscope is projected, by means of a prism, onto a screen, which is fixed in an adjustable tube at right angles to the optic axis, and can be viewed and focussed up to the moment of exposure. To insure accurate focus, the screen in the adjusting tube should be placed the same distance from the microscope as the plane of the sensitised plate. A slit in the shutter can be opened and closed to regulate the exposure.

A Clinical Device.—Dr. Eugene Shurtleff, of Boston, has simplified the clinical microscope so that everyone who has a draw tube with a society screw can have one. A sheet of brass three-fourth inches long and one-sixteenth thick is wide enough to nearly embrace the draw tube when bent. At the distal end of the sheet, two clips are cut out and beat so as to hold the slide. The coarse adjustment is easy; the fine adjustment is not so easy. Still, when once in focus, it is easy to keep it so. In quantities they can be furnished at twenty-five to fifty cents each.

An Improved Cell of Glass and Celluloid for the Preservation and Exhibition of Microscopic Eye-Spec-

imens.—C. A. Oliver in the *International Medical Magazine*, February, 1994, described an air-tight cell for the preservation of microscopic eye-specimens. It is made of two parts, the upper one being of glass in the shape of a petry, or chemical crystallizing dish, which sets in a celluloid base by means of a deep circular groove. The glass is filled with the preservative fluid (gelatine), the specimen introduced and the base applied, and the whole inverted, the raised bottom will press out all air bubbles and the glass can then be cemented to the base. A single hand magnifying glass of any amplification or the ordinary dissecting microscope is then used for examining the specimen.

The Jena Glasses.—The researches of Abbe and Schott at Jena, have resulted during the past 10 years in the production of several improved grades of optical glass, and especially the pairs of flint and crown glass in which the dispersion in various parts of the spectrum is much more nearly proportional. By the use of these glasses the magnitude of the secondary spectrum is greatly diminished. A combination of a heavy barium-phosphate-crown glass is made with a borate-flint glass in which the focus differences range between—0.04 and 0.79.

Erector for a Microscope.—In an ordinary terrestial telescope, the eye-piece consists of four lenses—two next the eye, which form a Huyghenian eye-piece, and two at a little distance from them, which perform the duty of reversing the image and enabling us to see it in its proper position. These two lenses will form an efficient erector for the microscope, or the whole four-lens eye-piece of the telescope may be used as an erecting eye-piece instead of the ordinary Huyghenian.—*Dr. Blacklock in The Eng. Mech.*

MICROSCOPICAL MANIPULATION.

Soldering Glass to Metal.—The piece of glass is first covered with a thin layer of platinum. This deposit is secured by brushing over the slightly heated glass a natural chloride of platinum mixed with essential oil of chamomile. The latter is slowly evaporated and when the white and odoriferous vapors have ceased, the temperature is raised to a red heat which

reduces the platinum and covers the tube with a layer of bright metal. Placing the metalized tube in a bath of sulphate of copper, and connecting the tube to the negative pole of a battery of suitable energy, there is deposited on the platinum a ring of copper. If the operation has been successful, the copper will be malleable and very adhesive. In this state a glass tube topped with copper can be treated like a genuine metallic tube and soldered to iron, copper, bronze, platinum, etc.—*Age of Steel.*

Formaldehyde for Hardening.—Dr. W. M. Eccles compares the hardening action of formic aldehyde on tissues with that of the fluids in general use. Absolute alcohol hardens most tissues in four or five days, but is apt to render them brittle or to harden them very unequally. Methylated spirit requires fourteen days. Chromic acid, with or without alcohol hardens in seven days, but renders the tissues brittle. Potassium bichromate in Muller's fluid requires six weeks and must be constantly renewed. Mercury perchloride hardens rapidly and interferes with subsequent staining.

But in the case of formic aldehyde, very soft varieties of normal and pathological structures, the most difficult to prepare, all can be well hardened in three days. None of the tissues became brittle, all being cut easily with an ether-freezing microtome after soaking in gum, and all staining well with logwood and eosine. At the same time the cells are unaltered in character or shape. For very soft tissues, use a 40 per cent solution, for harder ones, a 20 per cent solution will do, and for quite firm material, 10 per cent will do.—*Brit. Med. Jour.*

Writing upon Glass.—Take 2 parts shellac, 1 part Venice turpentine, and dissolve them in the water bath in 3 parts of oil of turpentine. After complete solution, 1 part lampblack is added and the solution well stirred.

MEDICAL MICROSCOPY.

Blood of Melancholia.—A microscopical examination of blood of twelve patients showed a marked diminution of globules and of hemoglobin. The administration of iron alone or combined with quinine or strychnine increased their numbers and produced corresponding improvement in the patients.

Concurrent Infections in Pulmonary Tuberculosis.—Dr. T. M. Prudden has published in the New York Medical Journal of July 7, 1894, an extremely interesting paper on some researches which he has made upon the above-named subject. Although his work is as yet incomplete there are strong indications that pulmonary tuberculosis is helped on very materially by the *Streptococcus pyogenes* when acting in conjunction with the *Bacillus tuberculosis.* This paper should be read by every physician. Perhaps Dr. Prudden, whose address is 437 W. 59th street, New York City, may have reprints for distribution. The paper is accompanied by 12 photo-micrographs which lend added interest to the subject. If his researches prove correct, we shall have an intelligible explanation of why consumptives are so much benefited by removal from cities and low altitudes to the mountains and to the high lands of the Rocky Mountain plateau. He has been led also to emphasize the need of antiseptic treatment, the avoidance of aerial infection and the giving of the most scrupulous attention to hygiene and sanitation. Space does not permit us to give an outline of the experiments performed with their results, but they are of the greatest interest to the medical microscopist.

Flies Carry Contagion.—The Egyptian physicians have decided that the gonorrhœal ophthalmia which there becomes epidemic in the hot months is carried by flies which are a perfect pest in Egypt. The flies have been noticed to feed greedily upon the secretions from sore eyes. Children only a few years old are overtaken with the disease when no other means of infection can be discerned than the operations of the flies.

Contributions to the Microscopic Anatomy of the Human Nasal Cavities, particularly of the Olfactory Mucous Membrane.—Suchannek (*Arch. of Otol.*, *XXII*, 4) recognizes in the human olfactory mucous membrane, in addition to the supporting fibres, typical olfactory and basal cells:

1. Genuine leucocytes, ninepin shaped and oval, with hyaline contents.

2. Cells which resemble leucocytes, but which proved to be cellular elements with a pedicle.

3. Transition cells from the pigmented "bell cells," having a small amount of pigmentation and hyaloid contents. He has

never seen this form except in cases of diabetes mellitus, and he leaves it an open question whether they are normal or pathological.

4. Distinctly pigmented, round, long or transversely oval cells, the longitudinal axis being generally parallel to the longitudinal axis of the rest of the epithelial cells.—*N. Y. M. J.*

The Microscope in Anchylostomiasis.—We are indebted to an Egyptian physician, Dr. M. Sandwith, of Cairo, for some interesting facts about this disease, in the diagnosis of which the microscope plays an important part. Anchylostomiasis is an insidious, wasting disease, characterized by progressive anæmia without apparent cause, and by digestive and nervous deterioration, occurring chiefly among earth and brick laborers of warm climates, caused by the presence in the duodenum of a blood-sucking nematode worm. It occasionally proves fatal but is capable of cure upon removal of the parasites, and it is capable of prevention by means of cleanliness. It is caused by the introduction into the stomach of a microscopical quantity of earth containing embryos of the parasite in its rhabdite form. The muddy water drank in Egypt often acts as a vehicle. Unwashed vegetables and unwashed hands of people who have handled the contaminated soil coming in contact with the mouth produce infection. A microscopical examination of water from 56 wells and ponds showed 16 to be infected. Continued exposure to the sun's heat during dry weather destroys the embryos.

But the eating of earth by the natives is the great and surest cause of infection. Suspecting this, Leichtenstern fed men with rhabdites and found eggs in their excrements a few weeks later. The worms themselves are not excreted.

The microscope furnishes the readiest means of detecting the parasite. In most cases eggs can be detected in a tiny portion of the suspected fæces. The fæces are placed in a test tube with a weak solution of carbolic acid and rendered fit for examination. Suspected material may be cultivated in damp earth exposed to the air, and if the ova are present worms will hatch out.

The urine is similar to that of ordinary anæmia, neutral or alkaline and of specific gravity 1010 to 1015. In advanced cases the microscope reveals a trace of albumen without casts.

An examination of the blood corpuscles in 173 cases showed some interesting data. In only three patients did the red corpuscles exceed 4,000,000; 23 per cent numbering over 3,000,000 to the cubic millimeter; 47 per cent over 2,000,000, and 28 per cent less than 2,000,000 red blood corpuscles.

The average gain of red corpuscles during treatment was 1,290,000. A boy of 18 years gained 2,542,000; another, aged 16, gained during one month 7 pounds weight and 2,208,000 red blood corpuscles. The corpuscles were counted by the Thoma-Zeiss method, taking the average of 6 to 10 squares. The average number of white corpuscles on admission was 10,360, and the average gain in hospital 5,370. The increase in hæmoglobin was from 22 to 32 per cent as shown by Gower's hæmoglobinometer.

Four gramme doses of thymol will often rid the patient of hundreds of worms. After from one to three doses the microscope usually fails to detect any eggs in the excreta. In feeble patients 25 grains of brandy are administered with the thymol. The excreta are disinfected with perchloride of mercury (1 in 500).

The microscope proved that the anæmia was best treated with a daily supply of one and a half grammes of sulphate of iron in water in three equal doses.

A very full and exceedingly interesting paper on this subject is found in the *Lancet* for June 2, 1894, pp. 1362–1368.

BACTERIOLOGY.

Removal of Pathogenic Bacteria from Drinking Water.—This is the title of a paper in the December *Technological Quarterly*, the author being Mr. Geo. W. Fuller, Biologist of the Lawrence Experiment Station in Massachusetts. He has found that filtration through sand removes the bacteria from water and renders it harmless. The results of his experiments with water infected with typhoid and other germs were extremely gratifying. His work, however, is largely confirmatory of European work. The Thames water has been filtered for several years and the average typhoid fever death-rate in London for 1886-'88 was only 1.6 per 10,000 inhabitants. In 1891, it was only 1.3 per 10,000, while that of Chicago was, in 1891, 16.0 or

twelve times as great. Even in Philadelphia, it was 6.4 per 10,-000. During 1892 Hamburg drank unfiltered water from the Elbe and had 17,975 cases of cholera, 7,611 of which proved fatal. Altona also drank Elbe water, but only after filtration; and it had 562 cases of cholera. Its population was about one-fourth that of Hamburg and its cases about one-fourteenth that of Hamburg. Many of those were importations from Hamburg and not of home production. Wandsbeck, a neighboring city, using sand-filtered water from an inland lake, got off with only 64 cases of cholera in its 20,000 inhabitants. In a street dividing Hamburg from Altona, the houses on one side were supplied with unfiltered water and had many cholera victims. On the opposite side, the people drank filtered water and mostly escaped cholera.

The Lawrence investigations, covering 5 years and including over 11,000 bacterial examinations of water, indicate the practicability of constructing filters which will economical remove over 99 per cent of the bacteria contained in the unfiltered water. The exact depth of sand required has been ascertained and the number of bacteria lodged in each depth of sand, viz: ¼ inch deep, 1,100,000 bacteria per grain; ½ inch deep, 320,000; 1 inch deep, 140,000; 2 inches deep, 21,000; 4 inches, 4,000; 6 inches, 1,600. Lawrence now has a filter, covering 2½ acres, which yields 5 million gallons of water daily. It is the only one of its kind in America.

BIOLOGICAL NOTES.

Staining Living Cells.—A live tadpole immersed in neutral red (1 to 2 300,000) absorbs the color so rapidly as to have some of its tissues stained in 15 minutes; and in a longer time all the tissues became perfectly red. The color not only penetrates the outer membranes, but would seem to be attracted by the cells. This color has been found to possess the greatest affinity for live cells of all the kinds of coloring matter.

DIATOMS.

To Fasten Arranged Diatoms.—Put a brush containing very weak gum-water at one end of the line of diatoms, when

the gum-water will run along the whole line of its own accord. When dry the diatoms will be fixed, but even then great care is required in putting on the cover-glass.—*No Sig.*

MICROSCOPICAL SOCIETIES.

Washington, D. C., L. M. Mooers, Secretary.

May 8, 1894.—This evening was held at the High School Building the 10th annual soiree. Dr. E. A. Balloch, a member of the committee of arrangements and an ex-President, introduced the speaker, Dr. E. A. Gibbs, who read the address of the President. The subject of the address was, "Some of the Uses of the Microscope," illustrated by stereopticon views. The apparatus employed in making photomicrographs by sun light and lamp light, was shown on the screen and explained.

The germs of tuberculosis, diphtheria and cholera were shown and attention called to the importance of a microscopical examination as an aid to the physician in making a prompt diagnosis. A short description of trichina spiralis was also given. Diatoms and Foramenifera showing the lower forms of vegetable and animal life, fat crystals of various oils, crossed ink lines showing "overflow" where both lines were wet, and lines of demarkation where the first was dry, and the application of same in detecting forgeries in writing, illustrated by a letter from a forged document, also a photograph of a promissory note in which the erasure of the words "or bearer" was the cause of a law suit.

With a microscope attachment to the stereopticon, the formation of crystals from solutions of copper sulphate and barium chloride was shown on the screen. A full list of objects shown was as follows:

1. Photography. Direct light. Lamp light.
2. Heliostat. Sun light.
3. Heliostat.
4. Photography. Opaque object. Lamp light.
5. Photography. Written matter.
6. Tubercle bacillus.
7. Diphtheria. (Klebs Loeffler bacillus). In membrane.
8. Diphtheria. Same from a culture.
9. Cholera spirillum or Koch's comma bacillus.
10. Colonies from air.

11. Trichina spiralis.
12. Diatoms. Arachnodiscus ehrenbergii.
13. " Triceratium distinctum.
14. " Same showing dots.
15. Diatoms. Terpsinæ musica.
16. Diatoms. Pleurosigma angulatum.
17. Foramenifera.

17½. Mimmulites laevigatus.

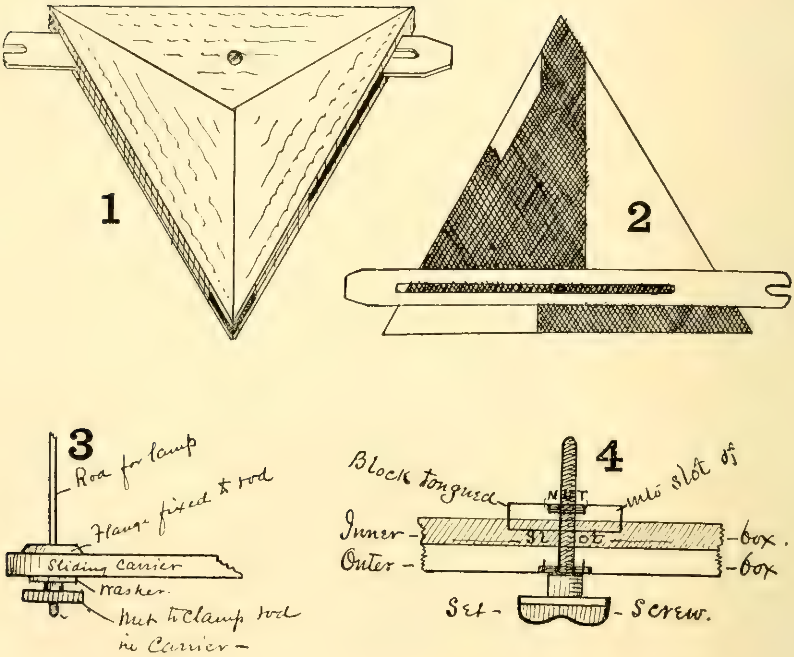
18. Fat Crystals. Cocoa nut fat.
19. Fat Crystals. Palm oil.
20. Fat Crystals. Carapa. Plain.
21. Fat Crystals. Chaulmugra oil.
22. Fat Crystals. Beef.
23. Fat Crystals. Butter.
24. Crossed ink lines, both wet.
25. Crossed ink lines, first dry.
26. An ink line and a pencil line.
27. Letter "J" from forged document.
28. Promissory Note.
29. Shows "e" in "Bearer" and cross line.
30. Shows "a" in "Bearer" and cross line.
31. Shows lines crossing "l" in "Charles."
32. Section of Oak stem. Crystals.

After the address, the exhibits by members were on view. Over 60 exhibits were made by 31 members, a full list of which may be found in *The Microscope* for April.

MICROSCOPICAL NOTES.

Vaccination is much more effective if practiced at once after recovery from typhoid fever, but no one knows why this is so.

Gallic and Tannic Acid Tests.—To a solution of tannic acid, add solution of chloride of barium and a pink precipitate will result, gradually darkening. To a solution of gallic acid, add a solution of potash and of chloride of barium and a blue precipitate will result.—*Fred'k Davis.*



DR. COUTANT'S SUPPORT FOR THE MICROSCOPE.

THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. AUGUST, 1894. No. 8.

A New Support for the Microscope.

By R. B. COUTANT, M. D.

TARRYTOWN, N. Y.

WITH FRONTISPIECE.

Every microscopist who uses his instrument in an upright position, has felt the need of a suitable support for it, one in which the problems of height, compactness, stability and the means of illumination are satisfactorily solved. No dealer in accessories supplies such an article and workers are forced to improvise one when occasion requires. An appliance specially devised for the purpose is seldom met with outside of well equipped laboratories and is not always to be found in them. The writer, after employing various make-shifts, invented the stand about to be described which answers all the purposes for which it was designed and besides is attractive in appearance.

The support is intended for the use of microscopes of large or small size in either an upright or an inclined position. It consists of two wooden boxes, triangular prisms in shape, one of which slides within the other and adds ten inches to the height of the stand when fully drawn out. Means are provided for securely fixing the movable box at any point within this limit. The stand is vertical in position and when in use is placed wedgewise between the legs of the observer, permitting a close approach to it and the use of an instrument in the erect position without fatigue.

The inner box supports a double top between the layers of which a lamp-carrier swings horizontally through an

arc of 90.° The carrier may also be lengthened or shortened according to need. The quadrant through which it moves corresponds with the left anterior corner of the support. The height of the boxes when closed is twenty-nine and one-half inches. The outer angles of the inner box are beveled so as to measure half an inch across, and strips of wood are set into the inner angles of the outer box so as to form sliding ways for the movements of the inner one, contact being at the angles only. The double, triangular top of the support is twelve and a half inches upon each of its sides and one inch thick. It projects three-fourths of an inch beyond the sides of the inner box so as to be flush with the top of the outer one, forming with the latter a cap to the support. Its upper and under layers are each made up of three triangular pieces of wood, (see Fig. 1) arranged so that the grain is parallel with the outer edges, to prevent warping and splitting. Each of the layers is one-third of an inch thick and the space between them for the sliding lamp-carrier measures the same. The layers are held apart by three pieces of wood placed as represented in the unshaded parts of figure 2, which also shows the mechanism of the lamp-carrier, the shaded area indicating space traversed by it in its movements. The piece of hickory of which the lamp-carrier is made is fifteen inches long, one and one-half inches wide and scant one-third of an inch thick. A slot half an inch wide and eight inches long is cut in the carrier, as shown in figure 2, and a screw which passes through the top of the support engages in the slot, guiding the movements of the carrier and hold it in place. The lamp used with the stand is the "Acme." A slot one inch long and three-eights of an inch wide is cut in the projecting end of the carrier, (see figures 1 and 2). In this, the upright rod of the lamp is fastened, as shown in figure 3, making it easy to fix and to remove the attachment. The lamp has universal

movements and may be raised or lowered, swung from side to front of stand, brought near to or removed from the mirror, inclined upon its axis, and rotated so that either the side or the edge of the wick is in use.

The outer box is eighteen and a half inches high including its feet. It is finished at the top with a rim of wood three-fourths of an inch wide and one inch deep, set flush with the inside of the box and attached to its edge by tongue and groove. There is a moulding around the base of the box three inches wide and one inch thick, to the projecting angles of which metal knobs are attached, forming a tripod upon which the stand rests. A large set-screw is fixed in the right hand panel of the outer box, five inches from the top. The corresponding side of the inner box is slotted, and the screw works through the opening, passing into a nut imbedded in a block which is tongued into the slot of the inner box. (Fig. 4.) When the screw is released the inner box may be raised or lowered: when it is tightened the sides of the boxes are securely clamped together and fixed at any elevation. The floor of the outer box is formed of a block of heavy wood four inches thick, which adds to the stability of the stand.

The boxes are made with three-eight-inch oak boards, thoroughly seasoned and arranged so as to counteract any tendency to warp. The inside angles are strengthened by triangular strips glued in and braded, and the layers of the top are secured in the same way. The outside surfaces of the boxes are polished and finished in spirit varnish.

The slot in the side of the inner box should extend from the bottom to within three inches of the top and be three-fourths of an inch wide. A frame of three-eights stuff, one inch wide, is fitted into the opening at the bottom of the box, bridging over the slot and stiffening the box where most needed. A portion of the frame

as large as the end of the sliding block, is cut out at the point where it crosses the slot, and a piece of metal is tacked over it below, making a recess in which the block is held when the set-screw is put in place.

The observer should be seated upon a chair the height of the stand when closed. The projecting ends of the lamp-carrier form handles by which the inner box may be lifted. The lamp and its attachments should be removed from the carrier when not in use.

Although primarily designed for the use of the microscope in an erect position, the stand has proved equally satisfactory with the instrument inclined. Indeed, for prolonged use by artificial light, it would be difficult to find a support by means of which so much of the wear upon the observer is done away with.

As the stand is not patented, anyone is at liberty to have it made, and it is the sincere wish of the writer that all who do so may find pleasure and profit in its use.

The Diatom Considered as a Protozoan, with Method of Demonstration.

By K. M. CUNNINGHAM,

MOBILE, ALA.

The article which appeared in the last issue of this periodical, entitled, "Studies in the Biology of the Diatoms," dealt more particularly with the motion, and epidermal covering of the diatom as presented by a number of species belonging to various genera. It has prepared the way for a further elucidation and study of the cause of their travelling movements, hitherto mysterious and puzzling.

Immediately on closing that paper, I resumed my verification studies of the various phenomena recorded in "A contribution to the Life history of the Diatomaceæ, by Prof. H. L. Smith," as referred in my preceding paper.

As all the results noted by myself, therein, were secured without the use of staining agents, or pigmentary colors, I conducted the new series of researches with the aid of water color pigments. I selected for the first trial, Prussian blue, in place of indigo. With a camel's hair brush, I transferred a small dab of color from the moist cake of blue, to the centre of a glass slip, and then with a rubber bulb tube, I deposited thereon a dip from a concentration of freshly gathered living diatoms. Placing the slide upon the stage, and adjusting a half inch lens to the tube, so as to secure a large field, I explored for specimens of active Navicula major, and having brought one to the center of the field, the half inch lens was changed for a sixth inch (Zeiss D) lens.

The N. major selected for study, offered its sutural face to the view. On confining my attention closely to its peripheral portions, I very promptly observed the characteristic molecular activity at the nodular cavities, and while reviewing these points, I witnessed in this single initial specimen (and so was enabled to corroborate and verify), all that had been previously described and figured by Prof. H. L. Smith in his paper. Finding that it was relatively easy to watch continuously all the phases of epidermal activity associated with the diatom, I set myself to master the character and causes of these varied living activities. And now after more than thirty days from the inception of the study with pigments and after devoting several hours each night of that period to observation of the living diatoms, I feel confident that the diatom will not offer further mystery as to its vital potentialities, except as to two absorbing metaphysical questions. These are: as to how its life may have originated and continues to manifest itself in the ordinarily hyaline protoplasmic matter characteristic of the initial beginnings of all life of which man has any cognizance; and second, the nature of the diatom's inherent chemico-

vital (metabolism) powers in its assimilation and elaboration of the various organic and inorganic elements requisite to the successful issue of its full life cycle.

To describe the character of material used in these determinations, I hope I may be excused for being somewhat prolix.

After having studied a special living material during a period of fifteen days—for a few hours each night, the material had become so filled with minute rotifers, that it necessitated securing a new supply of living diatoms. On the 4th of July, being urged on by the desire to solve the phenomenon noted by Prof. H. L. Smith and partially verified by myself, I visited an artificial fresh water lake at the Spring Hill College grounds, distant six miles from Mobile, from which locality I had secured the material at first studied. On arrival at the lake I found the lake used for bathing purposes and through the kindly aid of one of the bathers, I secured from the bottom of the lake a considerable tuft of eel-grass. I knew that the flat leaves were usually covered with living diatoms, and that they would be relatively free from sand and mud. This specimen was secured for the experimental supply of living diatoms, and while making tests outside of the lake, I found swamp-like spots filled with a reddish or iron-rust colored flocculent deposit. Making a trial test of this material for the presence of diatoms, I noted that it contained dead frustules in abundance. On account of their large and conspicuous sizes I collected a large mass of the vegetable growth and expressed the fluid therefrom into a bottle. It was wide mouthed, cylindrical in shape, an inch and a half in diameter and about four inches deep. I merely saved this material to see what species were present therein, as I did not expect to find living diatoms in the material. On returning to Mobile I submitted this material to microscopical examination, when I found that magnificent specimens of

the largest Naviculæ, such as N. nobilis, N. major, N. firma, N. rhomboides, Surirella splendida, Nitzschia sigma and Stauroneis gracilis, existed in the living state in connection with thousands of the empty frustules.

In all my previous years of experience in collecting diatoms from many sources, living as well as recent, or fossil, I had not found anything similar to this association of species. And perhaps, had it not been for this casual find, the solution of the mystery involved in the cause of motion in the diatoms, might have yet escaped what little biological or analytical skill I now possess, and I would have had to continue in the mental attitude of one who strongly suspects the animal character of the motile diatoms, but cannot crystallize his proof into a concrete expression for lack of demonstrable evidence. But this stage has been safely passed and an easy method of verification is accessible to all who use the microscope as an instrument of research, or for biological studies of any kind.

On the night of July 4, 1894, I determined that the bottle contained living diatoms, and set it aside until the evening of the 5th, but during the day, I found that the inner sides of both bottles of material, (the one from the eel-grass material of the lake, and the other from the swampy ground) were covered with thousands of living and moving diatoms that had come up out of the sediment at the bottom of the bottles. After having determined this, my uniform rule had been on each successive night to take the bottle containing the diatoms secured outside of the lake and twirl the bottle very gently to dislodge the living diatoms from the sides, taking care not to disturb the sediment at the bottom. After twirling the bottle, three-fourths of the clear water was poured into a flexible India rubber hemispherical cup about three inches in diameter. The material was then allowed to settle for two or three minutes, and the

water carefully poured back into the bottle, retaining in the cup about a half inch of water. The cup was then shaken alternately at right angles to cause the sediment to gather together, and was then finally twirled with a gentle circular motion, which massed the diatoms into a visible flocculent ball-like mass. At this stage more water can be poured off without disturbing the flocculent ball, when a bulb pippete will remove the diatoms with the least amount of liquid in the tube.

A drop from the tube is now deposited at the center of a glass slip. In the meantime, there should have been rubbed up a small quantity of India ink A drop of the India ink is added to the living diatoms, and a clean cover glass of at least three-fourth inch diameter is placed over the liquid. The slide is now placed on the stage of the microscope, and if inspection is made by means of half inch lens, it will be apparent at once, that all the living diatoms of the larger Navicula species are surrounded with either a narrow or broad peripheral hyaline or transparent zone. This is the evidence of the protoplasmic epidermis, and that the transparent and empty shells in most cases are not surrounded by the gelatinous coat. At other times the recently dead and partially transparent shells seem to be surrounded by the gelatinous layer, which of course is inert.

While the slide is under study two conditions may happen i.e., the layer of India ink may prove to be too dense, when nearly all transmitted light will be cut off, and the field appears as an opaque surface, though the diatoms will be partially visible as translucent spots. The other is when the layer of India ink presents a uniform layer of a grayish tone. Then the slide is proper for continued examination under a 1-6 lens or even higher powers. When the proper density of pigment obtains, the pseudociliar effect of the anterior, or advancing end of the protoplasmic mantle of the diatom is

readily recognized, and followed with a power of x 200, or a 1-2 inch lens, and A eye piece.

Attentive watching the marginal contact of the granules of carbon in advance of the path passed over will reveal the pseudopodial-like flow (characteristic of the movements of Amœba proteus) of the fringe of exoplasm at the advancing end of the diatom frustule. Then it may also be variously noted that the carbon area never comes into direct contact with the siliceous frustule, but seems to be driven away in advance and blown off to the sides of the diatom. It passes downward in the outer zone, or fringe exoplasm charged with the granules of carbon. If while the attention is thus occupied, and the diatom is seen to back, or take a retrograde movement, it will then be observed, that the end thus previously charged with the pseudopodial flow has become relatively passive and draws a long coma, or train of granules of carbon in its path. The current of carbon granules may be traced for considerable distance to the rear of the diatom. The carbon granules flow inward to the axial line of the diatom's motion after it passes any given visible stationary speck on the slide, but while the exoplasmic train of the retreating end of the diatom seems to be passive (i. e., not assisting in propulsion.) Yet from time to time, some particles of debris, or other matter may be gathered by it, and hurried into contact with the posterior end of the frustule. If the attention is now again directed to the anterior, or advancing end, the pseudopodial or amœba-like flow, or advanced undulations of the exoplasm will be again noted, as in the instance previously given. Should reversal of motion occur, the phenomena are repeated inversely as often as change of direction occurs.

Next the attention may be directed to the motility of particles of debris, flecks, or cloudlets of carbon driven along the peripheral edges of the frustule. The pseudo-

ciliary phenomena occuring alternately at both ends or prows of the Navicula species will not be so evident when studied under a 1-6 lens. For the penetrating power of the lens is very much reduced, when compared to the depth of layers of granules that the 1-2 inch lens will accommodate. For the appearance of granules of carbon rushing forward to contact with the frustule, or their apparent repulsion from the frustule, the 1-2 inch power is best adapted in order to demonstrate these various phases. The diatoms will appear relatively small, but the phenomena distinct, by using higher eyepieces without changing the 1-2 inch lens. The effects may be followed up to about 600 magnification.

But to witness the more interesting phases that compel a recognition of the evident Protozoan character of the Naviculæ, it is indispensible to use the highest powers possible. All the critical results of importance arrived at by myself were secured by the use of a Zeiss D lens, (equal to 1-6th,) A and B eyepieces, and a 1-2 inch Beck & Smith lens reversed in use, and adapted as an eyepiece. The latter combination resolved the test lines on Grammataphora marina, and some twenty-five or more species of Pleurosigma on a type plate of the Bay of Naples forms. The change from one eyepiece to another can be made at any opportune moment without loss of time, or changing the objective.

In regard to the lighting, and some other requisites of manipulation, an Argand burner lamp is used, a bull's-eye condenser being adjusted as near as possible to the flame, and a large image of the flame projected so as to fall on the concave face of the mirror. To the substage an achromatic condenser is adapted, and when the light is properly centered in the field, the result will be a dazzling light. But in order to guarantee the successful view of the various phenomena, it is necessary to have at hand a glass slip, or smaller piece of emerald or

grass-green colored glass (blue will not answer). This slip must be placed on top of the condenser, or the slide containing the living diatoms must rest directly upon the green glass slip. The utility of, and necessity for, the use of the green glass, arises from several causes. The green glass gives a quasi-monochromatic field, and limits all colors to black or white, and absorbs the chromatic rays, and more particularly the red rays so that one will find it impossible to recognize the true color of objects stained with any of the aniline stains, such as the red of fuschin, or violet, methyl blue, etc. The green glass intercepts the light from the mirror, but the green glass does not interfere in the slightest degree with the resolution of all lines or striations that the lens is capable of showing. The other valuable analytical feature is that the ciliary appendages of all infusorians from the minutest rotifer up are beautifully differentiated around their peripheries. This is indicated by a shimmering phosphorescent aureole surrounding the infusorians when their ciliary processes are active. This phosphorescent aureole is a prominent feature in determining the activity of the exoplasmic epiderm of the Navicula species. The slightest variation, or change of contour in any living particle that has a motile power proper to itself is thus readily made manifest, and only requires resolving power, combined with magnification, carried to the requisite degree with appropriate lenses.

With the modern Abbe condensers, various accesory modes of illumination are available in the study of the motile phenomena of the diatom, such as the use of oblique light, and central stop effects, in using the light. I have only been enabled to use central light in my studies. As a contrast to the evidence of ciliary action, as indicated by the phosphoresent aureole, if one should note and observe small particles of India ink under the same condition, no aureole of pale light would be seen

surrounding it, which fact would distinguish inert matter in some cases, from living matter on the slide.

When everything is in readiness as above outlined, a slide is prepared in the manner previously stated. If possible a single large living Navicula nobilis must be retained in the field of views. If it happens that the frustular face presenting the nodules and raphe are first shown, the attention may be directed attentively to the median nodule, when it is strongly probable that there will be seen at that point granules of India ink collecting and massing together, and rising upwards, then dispersed or carried away to the sides. At the same moment along the median line of the raphe an interrupted or continuous stream of granules of India ink will be seen traversing the raphe. As the granules retreat along the raphe, after having passed the nodular cavity, and when near the posterior or passive end of the diatom as then moving, the line of granules will be seen to be deflected from the raphe, and to pour into a cloudlet-like mass on one side of the passive end of the diatom. This phenomenon may be watched during the whole period of transit of the diatom. The action is sometimes intermittent and sometimes continuous for a short period; but no matter how long these motile appearances may be studied, a proper interpretation of them could not be readily had were it not for the fact that the diatom very often turns over from the valvular aspect (side view) to present its sutural face or front view, when the motile phases of the running carbon granules present an entirely different appearance.

This allows the phases anteriorly noted, to be duly interpreted, for as soon as, or very soon after the diatom turns over, there will be observed, inverted into the nodular cavities two solid lines of India ink granules, which are sometimes very sharply defined and are as clearly shown as if drawn with a pen and black ink on

white paper. It is partly from this phenomenon that the crucial proof of the Protozoan nature of the diatom must be drawn. During the nightly intervals for more than fifteen days that I studied these evidences of an animal nature, and at every time that I desired to study the "melanorreha," or black flow, in the living frustules, the phenomenon was always quickly exhibited if sought for, and might be followed without intermission for indefinite periods of study. To outline the whole range of molecular vital activity in the exoplasm of the diatom, and to indicate the full cycle of vital movements as seen, diagramatic sketches would be requisite. But in a general way the principal features may be outlined as follows: The living diatom (Navicula nobilis, major, viridis, divergens, or any of the larger Naviculæ), if seen in the field under the most favorable condition for study will show its Siliceous case, surrounded on both sides by a transparent or diaphanous zone of at least its own width or a little less. Around the exterior to the parallel zones is another width also equal to the width of the frustule. In this outer zone there is an appearance of great oscillating or vibrating activity in all granules used, whether of India ink, indigo, Prussian blue, carmine and bacteria, or any other microscopic granules that may be in the liquid. This oscillating activity seems to be due to the automotile power of the exoplasm itself, but at the same time the Brownian movement of particles everywhere else on the slide masks the interpretation of the sources of movement. But, as the external or outermost layer of the exoplasm has a distinct and undoubted segregating power over pigmentary granules, it may be safe to construe the eddyings and minute whirling vortices, bearing granules, as proper to the life function of the exoplasmic sheath. Apart from the zones thus indicated, there remains the alternating pseudopodial-flowing tissues surrounding the opposite or terminal prows of the

diatom, rendering service in an alternate manner as the diatom advances or retrogrades in its movements.

Should the formation of the sigmoid lines of black granules not be observed while the diatom is in motion, it will before long come to a halt. Then all at once the sigmoid lines will flash out and indicate their presence by a rapid flow of granules from the direction of the active end, flowing longitudinally towards the passive end. It will be noticed that the sigmoid lines dip into the depressions at the nodular cavities in sharply defined arcs of a circle without coming into contact with each other. After the flow of granules is lost in the nodular cavity, the granules from the first section of the sigmoid lines will be seen to emerge and flow towards the passive end of the frustule where they can be massed or dispersed by the exoplasmic fringe. This melanorrehic phenomenon may appear as unilateral or bilateral phases, the sigmoid lines having bilateral symmetry. These conditions seem to be governed by the instinctive will of the diatom, as in the case of the vorticellians, starting and arresting their ciliary whorls at their own apparent violition. These color current phases are oftenest seen while the frustule is at a standstill. When movement is again resumed, the currents flow inversely to the previous indications. To prepare for viewing these movements, it is necessary to focus carefully and exactly on the upper margins of the frustular edges when the sutural view or aspect is presented. For, when the valvular or lined face is shown, the current phenomena are not of sufficient significance or interest. These have already been detailed. While observing the granular flow on the sigmoid lines, it will be noted that they maintain a constant and fixed relation to the sides of the siliceous frustule for a specific period. A materially evident hyaline space separates the line of granules from the shell. It may be noted that the alate protoplasmic

tissues supporting or carrying the granules have a retractile and contractile power. Frequently, while being observed, the sigmoid line of granules may be expanded as if by a twitch-like movement to twice its former distance from the shell, since the sides of the siliceous frustule are strictly parallel. The estimation of the amount of expansive deflection of the sigmoid line of granules is very marked, and the deflection of the lines where they dip into and emerge from the nodular cavity is also very marked when the flow is active. Under favorable conditions the phenomenon of the pseudo-ciliary action is beautifully shown along the pleural fringes or zones. From the outer or second zone of exoplasmic activity, the full width of the fringe containing the active carbon granules may be seen to be rapidly cleared up by a spinning flow. This diverts all of the India ink granules to the extremity of a short arc-like thread that pours its current of granules into the nodular depression, and outwards again towards the passive end of the frustule. If the attention is now drawn to the anterior moving (active) prow, the pseudo-podial flow of the exoplasm (ectoderm) may be observed as long as desirable. This is closely analogous to that of an active moving amœba proteus or naked amœba. When merely moving without feeding, the ectoderm will seem to be bunched up like the advancing waves before the bow of a river steamboat and insinuating itself irregularly as well as expansively among the granules of carbon. This rapidly causes them to stream away to the right or left, and usually evenly parts them at the middle of its axial line of travel. The carbon granules do not infringe against the siliceous frustule unless drawn in there at the will of the diatom. The effect of the flowing, advancing exoplasmic mass is to act on the granules as if blown away, rather than as if driven off by ciliary action. The latter is readily noticed among the

smallest rotifers. They sometimes actively clear up a space around themselves five or more times the width of their own diameter, when the granules are seen to be projected with great rapidity. A one-sixth lens, having little penetration relatively, is in this case best adapted to showing the phases of the pseudopodial flow, rather than the apparent attraction or repulsion of the carbon granules. This, however, can be readily witnessed with a one-half inch lens as already described. In addition to the active and passive ends of the diatoms while in motion, there are other effects to be noted. Such are the peripheral wave motion or phosphorescent-like undulations of the exoplasm in contact with the siliceous sides of the frustule. When the sigmoid lines of granules are not visible, the play of the epidermic layers in the vicinity of the nodules (which are good tests for good eyesight), may be readily seen. Around these points are well shown the minute phosphorescent flashes of light when the emerald green glass is used as a stage screen to cut off the glare and red rays of the light.

A hypothetical conception of the tissues which control and support the sigmoid lines of granules is as follows. In the valular faces of the Naviculæ there is a slit-like line connecting the terminal nodules with the median nodular cavities of the Navicula. In this nodular cavity can always be seen the two dots indicating the extremities of the cleft of the raphe. Through these nodular dots, together with the cleft along the raphe, there is extruded at times an alate or ring-like process of protoplasm from the internal portion of the frustule. Its contractile and extensile phases may be controlled by some vital power resident in and appropriate to the functional power of the nucleus. This is situated in the endoplasm between the nodular expansions the same as in other nucleated organisms. When in action the alate protoplasmic appendages seem to possess an equivalent

firmness or stiffness of structure like the pseudopodia of any of the globose or villose amœba.

In certain large fossil specimens of Navicula nobilis, I have been enabled to recognize the siliceous raphe projecting above the valvular faces, and vanishing or tapering off on their entrance into the nodular cavity. This cavity is grossly evident in Navicula dariana, and clearly enough in all of the larger species of Navicula. In the dried and mounted, larger Naviculæ of these studies, the solid projections of whatever there may be of a raphe or keel thereon, does not show in the least in frustules showing their front (sutural) faces. Hence it may safely be assumed for the purpose of the hypothetical discussion of the same, that the raphoid alate projecting processes, are protoplasmic appendages of the diatom and that they vanish when the frustule dies, or is cleaned for mounting. All the leading motile phenomena referred to herein, have their corresponding analogies among the admitted or acknowledged Rhizopods and the Forameniferal members of the orders of the Protozoa.

In addition to the various motile aspects of the diatom in its sutural, non-travelling aspect, I will include two additional phases that I witnessed. I have seen a straight line of black pigment ejected at right angles to the nodule, and after a pause sucked back out of sight, the line of pigment being as long as twice the width of the frustule, and also a continuous ejection of the black current from a nodule and slowly coming up and away as a black close spiral. The coils widened as the ejection of pigment was kept up. But there was not going on at the same time the formation of the sigmoid lines of granules.

In the initial experiments with Prussian blue the rapid formation of drifts or cloudlets of snow-like tufts interfered with the inspection of the sigmoid lines, and resulted in my settling upon India ink for the final tests. In the material last used, I found living Navicula, after

twenty-two days inclusion in the experimental bottle. After immersion in the same water largely charged with 5 per cent of India ink for a period of either twenty-four or forty-eight hours, the nuclei of all living diatoms, N. nobilis, N. firma, etc., were very easily distinguished in their living condition. This was on or about the twentieth day of my studies; the N. firmas seemed to have absorbed the India ink around the globules and in the endochrome. The frustules at this time were densely packed with numerous globular masses and were quite dark. It is possible that the gelatine base of India ink is a food pabulum adapted to the diatoms, as it seems to agree with them while aniline stains which are toxic kill them.

Microscopical Technique Applied To Histology.—V.

[FROM THE FRENCH OF RENE BONEVAL.]

Continued from Vol. XV, page 213.

To Examine the Marrow and the Periosteum.—For the latter no special preparation is needed, sections made after decalcification of a piece from the periphery of a bone showing all the desirable details. To observe the elements of the marrow, break the femur of a young animal, take out a small piece of the marrow and dissociate it rapidly on a slide without fluid. Expose to osmic acid vapor, and stain in picro-carmine. . . .

STRIATED MUSCULAR TISSUE.

To study the sarcolemma, the nuclei, the fibrillæ, etc., which enter into the composition of muscular tissue, we should seek different animals because certain details of structure are more easily appreciable in one kind than in another. . . .

Sarcolemma.— Dissociate in water a bit of muscle from the sartorius of a frog; in a few minutes the water penetrates the sarcolemma by endosmose and sep-

arates it in places. An aqueous solution of iodine will make the membrane more apparent by coloring it yellow. Tear it off. We thus obtain fibres in which the striated substance is broken within the membrane and leaves, between the ends, a space where the sarcolemma is very apparent. (Formula for the solution of iodine: Water, 100; potassium iodide, 6; iodine, 4. Dilute with water if the stain is too intense.)

Muscular Fibres; Nuclei.—. . . .If a bit of muscle is put on a slide with a drop of the 40 per cent potash solution, a slight teasing with needles will give admirably isolated fibres. Examine in the same liquid as they cannot be preserved. Permanent preparations are made after maceration in the ⅓ alcohol. Skin a frog and put a hind leg in the ⅓ alcohol. In 24 hours remove a bit of the sartorius, and dissociate on a slide by partial drying. Stain with picro-carmine for ten minutes; apply the thin cover, and wash gently by running a drop or two of water under the cover, replacing it by acetic or formic acid glycerine.

Muscular Fibrils.—The fibrils of insects show, in an exceedingly elegant way, the transverse striation of the contractile substance. Select a beetle and remove the elytra and the wings. Cut open the thorax so as to expose the wing muscles which will appear as a whitish mass. Place in the 1-3 alcohol for 24 hours. With curved scissors cut off a bit and dissociate it on a slide by partial drying. Stain deeply by an old preparation of hæmatoxylin. Mount in balsam.

Sections.—. . . .Put a frog's hind leg in a 2 per cent bichromate solution. After 10 days remove the sartorius muscle, wash, hardened in alcohol, embed in gum. Transverse sections are the most instructive. Stain by hæmatoxylin and eosine; mount in balsam. . . .It is necessary to section also the muscle of a mammal as the arrangement of the nuclei is different.

Bowmann's Disks.—Macerate a small muscle in dilute acetic acid (1 c.c. acetic acid to 400 grms. water,) and dissociate on a slide by needles. . . .

Vessels of Striated Muscles.—Inject an animal with the Prussian blue gelatine to be described hereafter ; choose a muscle with parallel fibres, and make longitudinal and transverse sections after fixing in bichromate, hardening in alcohol, embedding in gum ; mount in balsam. Instead of sectioning we can take a thin and transparent abdominal muscle of a frog, spread it on a slide and mount it flat in balsam.

Nerve Endings in Muscle.—. . . .From a frog just killed take a muscle and fasten it to cork by sharp quills. For five minutes place it in lemon juice freshly expressed and filtered through flannel ; wash the muscle rapidly and put it in a 1 per cent solution of chloride of gold for 20 minutes. Wash again for one second, reduce in the dark in $\frac{1}{4}$ formic acid (water, 4 ; formic acid 1). The next day, the reduction of the gold completed, mount in formic acid glycerine. . . .

To study the Motor Plates of the Lizard.—Mix 4 parts of chloride of gold with one part of formic acid. Boil and cool. Place a bit of muscle in 2 c. c. of pure gold chloride and add drop by drop a few c. c. of the boiled gold and acid. Reduce in the $\frac{1}{4}$ acid. Dissociate on a slide ; mount in formic acid glycerine.. . . .

UNSTRIATED MUSCULAR TISSUE.

For this study nothing is better than a frog's bladder. Kill a frog and put a strong ligature around the cloacal orifice. Open the abdomen on the lower surface and cut the rectum at the upper end. In the rectum insert a syringe charged with the 1-3 alcohol, or with ammonia bichromate, and gently depress the piston. The fluid enters the cloaca where the ligature retains and forces it to pass into and to distend the bladder. . . .Place a ligature beyond the syringe, remove in one piece the

bladder, the rectum and the posterior members, and put the whole till the next day in the fluid used for the injection. Remove the distended bladder, slit it open under water, and with the internal surface upward brush it until all the epithelium is washed away. Spread it, internal face upward, on a slide, and partly dry it. Stain with hæmatoxylin and eosin; mount in balsam.

Dissociation of Unstriated Tissue.—The foregoing preparation will show the muscle cells almost completely isolated, as well as cells united in fibres, yet it is important to continue the observation by other methods.

With curved scissors remove a bit of the muscular layer of the intestine and put it in a drop of the 40 per cent caustic potash solution. In a few minutes dissociate with needles; cover and examine in the potash. This preparation will not keep.

To make permanent preparations of dissociated muscle cells, proceed thus. With a ligature close a loop of the small intestine of a rabbit, and remove it. Into it inject a 2 per cent solution of ammonia bichromate. When sufficiently distended tie it beyond the syringe, and put it for 1 or 2 days in a bath of the same solution. Wash for 24 hours, after renewing the water. With forceps a shred of the muscular layer is easily torn off. Dissociate it in a drop of water on a slide. Stain with picro-carmine in a moist chamber for 12 hours; mount in glycerine. To observe the nuclei, stain with alum carmine for 1 or 2 hours instead of with picro-carmine.

Vessels in unstriated muscles.—From a rabbit whose blood vessels have been injected with Prussian blue, remove a portion of the small intestine and put it in a 2 per cent solution of ammonia bichromate. After 1 or 2 days' maceration remove by scalpel and forceps as thin a piece as possible from the muscular layer. Wash, stain with hæmatoxylin, mount in balsam.

Sections of Unstriated Muscles.—Fix an intestinal loop

in strong alcohol, after injecting it with the alcohol as just described. Imbed in gum, stain with picro-carmine, mount in glycerine.

Nerves of unstriated muscles.—The frog's bladder should be used to show the nerve endings in unstriated muscles. Distend the bladder (as already described) with the boiled gold chloride and formic acid. Tie, remove the bladder, and put it for 25 minutes in the gold solution. Then open the bladder, wash, and reduce the gold in darkness for 24 hours in the ¼ formic acid. Extend with the inner surface upward, brush gently to remove the epithelium, and mount in formic acid glycerine. . . .

THE LYMPH.

The simplest way to collect the lymph is to take it from the dorsal lymphatic sac of a frog by means of a fine pointed pipette. Select a vigorous frog, fasten it by the feet and carefully remove the skin of the back. With a finger of the left hand press on the dorsal region to force the lymph to a determined spot. By a quick movement thrust in the point of the pipette which will partly fill itself ; more may be sucked up. The frog may be curarized, and the lymph taken from the dorsal sac. By curarization the lymphatic sacs are filled by such a quantity of lymph that the tongue hangs out of the mouth. To obtain the lymph, prick the sac with a fine pointed pipette.

To prepare a drop of lymph.—. . . .Put a drop on a slide. Expose for 5 minutes to osmic acid vapor, stain with alum carmine, cover, mount in neutral glycerine which is allowed to run under slowly to replace the alum carmine. This preparation shows in a remarkable way the varied forms of nuclei. If the action of the osmium is prolonged, the fatty granules in the white corpuscles will be blackened.

Put a drop of lymph on a slide, cover, and allow the

iodine solution to run under. This stains the lymphatic globules mahogany brown. . . .

Globules examined in the serum appear uniformly granular and the nuclei are invisible. Run under a little water; the protoplasm swells, becomes transparent, the nuclei appear and exhibit their whimsical forms.

To examine the migrating ability of the white corpuscles.—. . . .Insert a small cylinder of elder pith into the dorsal sac of a frog. Let it remain 24 hours; expose to the vapor of osmic acid. Cut longitudinal sections, and examine in water. To make the preparation permanent, stain in alum carmine and mount in glycerine. Only the cells in the superficial layers show amœboid expansions, those in the central parts are rounded and have undergone fatty degeneration; these are dead elements, analgous to pus globules. This experiment proves that oxygen is necessary for the amœboid activity of lymphatic cells; where this gas is absent (in the centre of the pith stick), the amœboid activity ceases, and the cell dies.

To observe the absorption of granules.—. . . .Rub up in a little water some vermillion or an aniline blue insoluble in water, until the coloring matter is an impalpable powder. Inject into a frog's dorsal sac 1 c. c. of water to which has been added a small quantity of this mixture. In a few hours the lymph cells of the dorsal sac contain granules of the coloring matter. . . .

THE BLOOD.

. . . .A few drops are enough for microscopical examination. Prick the compressed finger, the ear of a rabbit or the pads of a dog's foot; take frog's blood directly from the heart, otherwise it will contain a considerable amount of lymph.

To examine living blood.—. . . .Place a drop on a slide, cover, and add a ring of paraffin to prevent evaporation. Do this with blood of mammals and of frogs to show the different shape of the red corpuscles. In a few minutes

the corpuscles will form rouleaux like piles of coins. After a time many globules lose the circular form, the margins becoming irregular, or "crenated". . . .

To be continued.

EDITORIAL.

The Brownian Movement.

What it is.—When certain very minute particles of matter either organic or inorganic are held in a state of suspension in fluids, very active movements of the particles may be seen under the microscope. Gamboge has been often recommended for the purpose. But of all substances, carmine made of cochineal seems to be capable of the finest division and of the most brilliant illumination, and hence best for the purpose. The solution in water must be weak. By daylight on a back ground of faint blue and by lamp light on a golden back ground, thousands of tiny particles may be seen bright as rubies and moving about over the whole field of view. The smaller the particles the more vivid the movements. In alcohol, fixed and volatile oils, this movement is not observable.

The Discoverer.—The one who first called attention to it in 1827, was Dr. Robert Brown, a distinguished botanist. His paper was entitled : " A brief account of microscopical observations made in the months of June, July and August, 1827, on the particles contained in the pollen of plants, and on the general existence of active molecules in organic and inorganic bodies."

About a year latter he summed up his results as follows: That extremely minute particles of solid matter whether obtained from organic or inorganic substances, when suspended in pure water, or in some other aqueous fluids exhibit motions for which I am unable to account, and which, from their irregularity and seeming independence, resemble in a remarkable degree the less rapid motions of some of the simplest animalcules of infusions. That the smallest moving particles observed appear to be spherical or nearly so, and to be between 1-20,000 and 1-30,000 of an inch in diameter, and that other particles of greater and various size, either of similar or very

different figure also present analogous motions in like circumstances.

Many Causes Suggested.—Among the surmises that have been made may be mentioned: (1) Gravitation or the attraction and repulsion of the particles among themselves. (2) Currents contained in the fluids. (3) Evaporation of the containing fluids. (4) Energy derived from without the fluids, such as light, heat, electricity, magnetism. (5) The molecular motion of the particles of the fluid itself.

Gravitation and Currents Insufficient.—R. M. Bache has shown this in saying:

"Just as one sees a boat managed by an unskillful helmsman pursue its erratic way in going about, being taken aback, or heeled over by a flaw of wind, without for a moment attributing its movement to currents or any other cause but the true one, so the constant observer of the brownian movements knows full well that the particles themselves are moving, not being moved by currents or by gravitation towards the earth or among themselves. He, from the first, recognizes the fact that the smaller the particles are, the more vivid is their movement. He recognizes another, that, although many large particles do not, as masses, move at all, yet the larger masses are all alive, as it were, with smaller ones, seen clearly around their periphery, on the silhouette of which they are seen plying like banks of oars in an ancient trireme. He is struck with and convinced of still another thing, that whereas one might expect to find that all particles would manifest an attraction for each other through gravitation, and that the larger and largest, but all in proportion to their relative size, would attract and absorb the relatively smaller and smallest ones, nothing of the kind occurs, but the smaller, down to the smallest, go their own way, sometimes even touching the largest and bounding off and away as if they do not, as indeed they do not visibly, submit to the force of gravitation. Of course they cannot escape the influence of gravitation, whether terrestrial or among themselves, but the effect of gravitation upon them is masked, in what manner will appear latter."

Magnetism Insufficient.—Bache says:

"It seemed to me that magnetic earth-waves might affect particles in such delicate suspension as those of which we are

speaking, some of which are no greater in diameter than 1-100,-000 of an inch, seen under various powers capable of magnifying from 650 to 1300 diameters. Accordingly, I have placed the particular fluid under examination in the lines of force of a permanent magnet, with the magnet on one side and the keeper on the other of the drop of fluid. Concentrating the gaze on individual particles, to observe if their movement were modified, and then on others in succession, and often repeating the experiment, nothing could be observed other than the movements existing before the magnet had been brought into requisition. The only kind of particles susceptible to the influence of the magnet were those of precipitated iron, but iron is always obedient to the magnet."

Heat and Cold Insufficient.—Bache reports:

"Heat I applied in various ways, either irregularly or in an endeavor to distribute it as equally as possible on the glass slide on which the particular experiment was made. Mere currents are set up during the adjustment of temperature from radiation. At the same time one can observe and differentiate the motions due to the Brownian movements, the motions along currents, and also the motions from terrestrial gravity, if any, exhibited by particles, if the specific gravity of the substance be great, and the microscope be set at an angle with the vertical.

Cold I also applied, putting the slides with their cover glasses in a freezing mixture of broken ice and water, and reducing them to a very low temperature. Still the movements went on as apparently unmodified as ever. Herr Exner says, that glycerin, which under ordinary conditions shows absolutely none, or almost no molecular movement, shows it clearly when warmed up to the temperature of fifty degrees centigrade. In all the finely divided bodies, however, which I examined, there seemed to be no increase or diminution in the intensity of the movements, corresponding with their alternate subjection to heat and cold. There were occasions in which I thought that I observed acceleration from light, but I always ended by imputing it to the force of imagination, and if it were not justly ascribable to that cause, the fact that it could be so ascribed, is proof positive that if, through the influence of light and heat, any intensification of the movements of the particles took place,

it must have been very small. Moreover, the evidence is certainly here, to show that even if the movement were intensified by light or heat, that was the only influence that could be ascribed to them, that light and heat could not be deemed the cause of the movement. And lastly, Herr Wiener's micrometric measurements of the range of movement at different temperatures completely bore out this conclusion."

The Red Wave of Light.—The opinion of Bache is this:

"The theory of Herr Weiner, that the movements are due to the action of the red-wave of light and heat is refuted by the single fact that, as may be proved by experiment, one may interpose at pleasure·between the source of light or heat and the particles, either a violet glass or a red glass, without being able to observe the slightest alteration in the movements, either as to their range or their velocity. That is to say, red rays may be either partially excluded or selectively admitted, without diminishing or increasing the liveliness of movement. Hence light can have nothing to do with the phenomenon under discussion."

Electric Currents.—The galvanic current has been passed through liquids filled with particles without the slightest visible effect upon them.

Evaporation.—It was conceived that evaporation might be accompanied with a series of minute explosions which produce shocks that could manifest themselves through the mass of an aqueous solution, in the form of minute movements of finely divided matter held in instable equilibrium by suspension in the fluid. Bache had satisfactorily eliminated nearly all other hypotheses and held this one for final examination. He says:

"At this point I encountered an obstacle. My high powers of the microscope were both water-immersion lenses. It seemed, therefore, that even when I had had the drop of liquid under observation, sealed beneath a cover glass, I might have included, by the use of the water-immersion lens itself, an evaporating surface which might have produced the optical illusion of the movement of the particles in suspension. I proceeded, however, with my experiments, upon the assumption that this, as the event proved to be the case, was not true, and meanwhile procured from Vienna a one-fifteenth dry lens by Reichert, the highest power of dry lens that he makes,

"I had already obtained for high-power lenses a film of liquid thin enough to be observed through all its strata, free of air within the cell, and protected from evaporation by being hermetically sealed. Any ordinary manufactured cell is too deep, and with all precautions taken contains a little air. On the other hand, the mere cover-glass superposed on a glass slide contains too slight a depth of fluid. I made a cell by using gum-shellac traced in a circlet on a glass slide, which cell, when partially dried, is filled to the brim with the liquid to be observed upon, whereupon the cover-glass is pressed into the yielding gum, thereby expressing the contained air with the superfluous liquid, when the product, dried over night, is fit for use on the following evening. One slide, prepared in this manner and filled with a slightly tinted solution of carmine from cochineal, had been observed upon by me for weeks, with a one-tenth water-immersion lens, and afterwards, upon the arrival of the one-fifteenth dry lens, was observed upon without showing any variation in the range and vividness of movement of the particles subjected to examination. I have even covered the whole microscope with a pall of thick, black, woolen cloth, so that not a ray of light could enter it, either through the cover glass or the eye-piece, and then carefully placing the eye close to the eye-piece, have suddenly thrown light upon the cover-glass, when the Brownian movement among the particles was perceived in as active play as ever. I have, therefore, concluded, from all these experiments, that neither heat nor light, nor electricity, nor magnetism, nor mechanical shock, nor finally evaporation, is operative in producing the movements; in a word, that the particles move uninfluenced by these forces."

The Fluid Moves the Particles.—All other possible suppositions having been eliminated, Bache has concluded that it is not the particles which are moved by their own energy, or moved by any energy directly imparted to them from outside sources, but that it is the fluid that moves them. Accordingly he argues as follows:

"If their own energy moves the particles, we should see them at the same time obedient also to the law of gravitation among themselves, manifested as the resultant of whatever forces are in play, whereas, although they must be obedient to the law of gravitation among themselves, its effects, and generally, as

well, those of terrestrial gravity, are so masked as not to be at all perceptible. Now, when we consider how minute all of these particles are, and yet that they move apparently unhindered with such constancy and force, it ought to be apparent that they have no self-motive power. However erratic the paths of individual particles may be, the likeness among the movements is extraordinary, so almost identical in every case, varying in greatness of range and rapidity only in inverse ratio to the size of the particle, that we cannot conceive of self-actuated particles so behaving; for relative greatness of size in self-actuated particles ought to coincide with relatively greater, not relatively less, energy of movement; whereas, here the case is reversed. But there are other facts that I have observed through experiment, which also prove what I say. In alcohol, and as far as my experiments go, in fixed and volatile oils, the Brownian movements are not observable, and yet the microscope plainly reveals that the movements of foreign bodies in all these is as free as in aqueous solutions, and I think more so. So molecular movements of solid particles in suspension in aqueous fluids must take place perforce of the constant repulsions of the constituent molecules of the particular liquid present—water."

"I have shown that the molecular motion, called Brownian, taking place under all conditions imposable, is a property of water and of water only, and that light and heat have naught to do with producing it, although, as I have admitted, they may possibly act in intensifying it. All that I may claim to have detected is a phenomenon which reverts to the molecular constitution of water, as to which the moving, solid particles in it concerned in the brownian movement have no more to do than has a current-metre to do with the flow of the stream the swiftness of which it measures."

Attention is thus fully called at this time to the latest explanations of the Brownian movements, in order that they may be considered in connection with the very interesting paper of Mr. Cunningham upon the movements of diatoms.

Washing Diatoms.—To avoid the use of acid in clearing diatoms a correspondent of a country publication says, put it in a jar of water and shake it for *an hour*. Evidently the time of this man is of little value and the space of the editor equally so.

MICROSCOPICAL MANIPULATION.

An Experiment.—Take some dust from the gutter of your roof and put it in a little tank of water. Examine it under the microscope daily. Perhaps you will find a Rotifer, an Amœba or an Infusoria.

To Mount a Leaf of Deutzia.—Boil in nitric acid and chlorate of potash until the leaf is tender. Then take off the epidermis with needles, stain it with hæmatoxylin. After washing, dehydrate it with alcohol. Clean it with oil of cloves. Mount in balsam.

DIATOMS.

To give some idea who have been the great writers upon the diatoms we have counted up the number of articles by each cited in the bibliography which is attached to Mills' Introduction to the study of the Diatomaceæ. We omit those who are cited less than ten times.

C. G. Ehrenberg, Berlin.	152
A. Grunow, Anvers.	102
Conte Astracane, Rome.	81
Rev. E. O'Meara, London.	63
Fred Kitton, London.	60
G. B. DeToni, Venezia.	40
Paul Petit, Paris.	33
Julian Deby, England.	32
H. L. Smith, Geneva, N. Y.	32
J. W. Bailey, New Haven, Conn.	32
L. Rabenhorst, Dresden.	28
K. K. Greville, Edinburg.	26
A. M. Edwards, Newark, N. J.	26
H. Van Heurck, Brussels.	25
J. Pelletan, Paris.	25
Matteo Dott Lanzi, Rome.	21
E. M. Nelson, London.	19
Charles Stodder, Boston.	18
Otto Muller, Berlin.	16
W. Gregory, M. D., London.	15

P. T. Cleve, Paris.	14
G. C. Wallich, London.	13
Dr. F. Cohn, Breslau.	13
Dr. J. F. Weiss, St. Petersburg.	13
G. A. W. Arnot, London.	13
J. Brun, Brussels.	13
Hon. J. D Cox, Cincinnati, Ohio.	12
L. Dipple, Germany.	12
J. J. Woodward, Washington.	12
C. H. Kain, Camden, N. J.	11
G. Schaarschmidt Jstvanffi, Klausenberg,	11

NEW PUBLICATIONS.

Sewage Disposal in the United States.—By George W. Rafter, and M. N. Baker., New York. D. Van Nostrand Co. $6.00. 2nd edition ; pp. 598, 102 figures.

Among the gigantic problems of city life is that of sewage disposal. The sciences of Engineering, Chemistry and Biology are called upon to do their utmost in this direction. The literature of the subject is already voluminous and will constantly increase, but nowhere will be found a more valuable contribution than that under notice. Its distinguishing novelty and its feature of most interest to us, is the study of minute organic life with the microscope with application to the sewage problem.

We now know beyond peradventure that the germs of Typhoid, Cholera, Dysentery and Diarrhœa get into sewage and therefrom pass at times to drinking water to be again admitted to the system with deadly effect. How to prevent this cause of epidemics is a problem treated by this volume, but we cannot take space to go into details. A full and absorbing history of many epidemics is given with the causes clearly traced.

Some microscopic forms of a vegetable character are injurious to drinking water, while some others have a purifying effect upon sewage laden streams. To the inquirer as to the utility of microscopical study, a very satisfactory answer can be made under this heading. Attention may be directed to the chapter on: "Self-purification of a running stream from a bio-

logical point of view." Perhaps it is not too much to hope that such men as Rafter will yet find out just what animals and plants may be used to purify sewage laden streams and how to rear such animals and plants in enormous numbers.

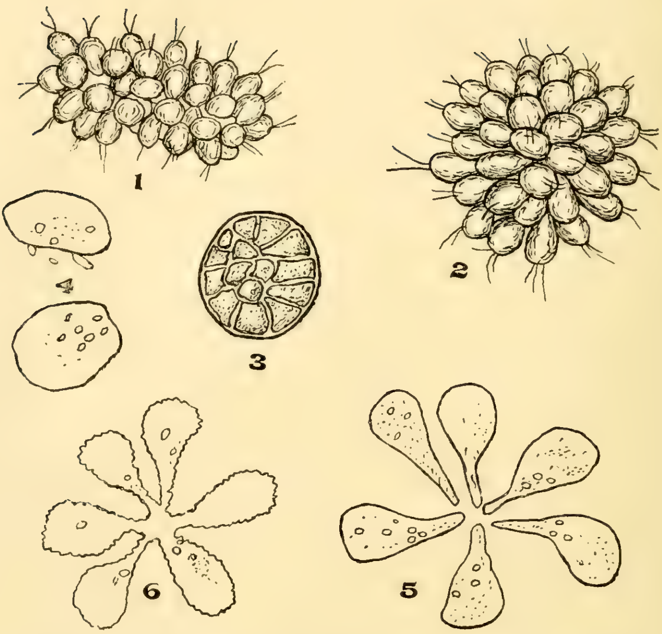
Among the hopeful animals is *Paramecium aurelia*, an animal about 1-90 inch in length. It is a filth infusorian, but an interesting form for study. Twenty to fifty may often be found in a single c.c. of contaminated water. A bright green colored infusorian, *Euglena viridis*, only 1-500 inch in length is often found in standing water. As it multiplies by fission and not sexually, enormous numbers can be produced in a short time. Hydra and the rotifers are also found in bad water. The precise function of each organism is yet to be worked out, though we believe in a general way that they purify the water.

Entomostraca eat dead animal matter, can be kept alive for months on human excreta and will soon die without it. They in turn constitute an important food for fishes. Those Fish commissioners who are blind to these facts, and most of them are so, are neglecting perhaps the most important topic within their reach. Nature undoubtedly has the requisite agencies for performing economically all these transformations from the worst of filth to healthy organisms. We must wrest from her the secrets of this work.

The diatoms come in for notice also. In one specimen of water, Rafter found 1792 *Synedra*. Is their mission healthful? Of Algæ, 2664 zoospores were found in one specimen.

The value of sewage as a fertilizer is declared to be too small for commercial consideration on account of the very great dilution. Comparison is made with a pint of brandy mixed with a hogshead of water, or the minute particles of gold contained in the earth upon which Philadelphia is built. In the aggregate there is said to be a thousand million dollars worth of gold in the bed of clay in question, but the cost of extracting it would far exceed the product.

It is impossible to give an idea of the immense storehouse of knowledge contained in this volume every page of which is strewn with facts bearing upon the problem. The maps, and other illustrations have been prepared at great expense. The price of the book is high, but city and town authorities must have it in the study of sewage disposal.



SYNURA.—A PLANT INJURIOUS TO DRINKING WATER.

Fig. 1.—Elongated colony.

Fig. 2.—Spherical colony—ordinary form.

Fig. 3.—Encysted state.

Fig. 4.—Disintegrating zooids—oil globules escaping.

Fig. 5.—Zooids flattened under the cover-glass, showing oil-globules.

Fig. 6.—Zooids flattened under the cover-glass—a variety with crenulated margin. Not common.

THE AMERICAN
MONTHLY
MICROSCOPICAL JOURNAL.

VOL. XV. SEPTEMBER, 1894. No. 9.

Synura.

By GEORGE C. WHIPPLE,
BOSTON, MASS.

[See 18th Ann. Rep. Boston Water Board, 1893.]

Of the thirty or more genera of infusoria which are found in the water supplies of Massachusetts, there are but fifteen which may be said to be commonly found in large numbers. Eight of these common forms belong to one order, and six of them belong to one family of that order, if we adopt the classification of Mr. W. Saville Kent.*

The special characteristic of this family, the Chrysomonadidæ, is the presence of lateral pigment bands. These color bands, in addition to their distinctive tint, are apparently of firmer consistency than the surrounding transparent protoplasm, and bear a very considerable resemblance to the coloring matter of the Diatomaceæ."

But the most important fact about the Chrysomonadidæ, from a sanitary point of view, is that almost every one of them has given rise to very disagreeable and sometimes extremely offensive tasts and odors in the waters in which they have been found. Uroglena, Cryptomonas, and Chloromonas have already acquired quite unenviable reputations. To these may be added Synura uvella and Dinobryon. It is noticed also that there is a similarity between the tastes produced by some of the organisms of this group and those produced by certain diatoms. Cryptomonas, for instance, produces a sweet-

* A manual of the Infusoria, I, 212.

ish, aromatic taste, very much like that of the violet. The diatom Asterionella also produces a sweetish, aromatic taste and odor resembling that of a rose geranium, although at times the Asterionella odor is decidedly fishy and oily. Uroglena volvox has a strong oily taste, very much like cod-liver oil. Synura uvella has, at times, a somewhat oily taste, often resembling that of a cucumber, but generally more spicy and bitter. The taste is a very persistent one. "It stays in the mouth." It is strongest at the base of the tongue, where the nerves are most sensitive to bitter substances. The taste of Dinobryon is similar to that of Synura, but is not as strong. In all of the above mentioned organisms oil-globules have been observed. In some of them the amount of oil has been estimated, and in at least one of them, Uroglena americana,* the oil has been isolated. It remains to be determined if there is any connection between the presence of the pigment bands and the amount of oil production.

It should be stated that these organisms do not always contain oil-globules. In the younger forms they are frequently absent. The oil may be said to be a reserve product, produced by the organism during its growth, and stored up in the cell,—hence it is most common in the older specimens. It is by the disintegration of the cells and the consequent liberation of the oil that the tastes are brought about.

"The Synura animalcules are free-swimming, united in sub-spherical, elongated, social clusters, each zooid contained in a separate membranous sheath or lorica, the posterior extremities of which are confluent. The contained animalcules almost entirely fill the cavities of the loricæ, their posterior extremities being produced towards and adherent to the bottom of the sam.e The

* See "Odors in Drinking Water" by Gary N. Calkins in 24th Ann. Rep. Mass. State Board of Health, 1892.

two flagella are sub-equal. Minute eye-like pigment-specks are sometimes present, though generally absent. A large vacuolar space, apparently representing a pharyngeal dilatation, is developed at the anterior extremity. The yellowish-brown color bands are produced equally throughout the length of the two lateral borders. The contractile vesicles are two or three in number, posteriorly located."*

The size of the colonies varies from 30 to 75 microns in diameter. Generally there are about twenty zooids in a colony, though sometimes there are as many as forty. The spherical colonies are often seen moving briskly through the water with a rolling motion. The elongated forms generally move more slowly. At a certain stage in its life history, Synura becomes encysted. In this condition it is smaller in size, and the zooids are crowded together and surrounded by a sheath. It is also somewhat darker in color, and is entirely without motion.

Synura in its maturer condition contains oil-globules. They are especially numerous just before encystment.

At times the amount of oil has been approximately determined. On December 9, 1893, a sample from Basin 3 of the Boston water supply contained 100 colonies of Synura per c.c. It had a strong, bitter taste. Each colony had about 20 zooids, and each zooid contained about 20 oil-globules which had an average size of about one cubic micron. Calculation showed that oil was present approximately in the proportion of one part of oil to 25,000,000 parts of water. This seems to be a very small quantity of oil to produce so strong a taste, but some experiments on a few of the essential oils prove that it is easily within the range of possibility.

The following table shows the degree of dilution at

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* See Kent, loc. cit., I, 412.

which some of the essential oils can be recognized by tasts.

Oil of peppermint	1 : 50,000,000
Oil of cloves	1 : 8,000,000
Oil of checkerberry	1 : 7,000,000
Oil of cassia	1 : 6,250,000
Oil of bergamot	1 : 6,250,000
Cod-liver oil	1 : 1,000,000
Kerosene oil	1 : 800,000

Synura is generally found in surface waters where there is a considerable quantity of organic matter. It does not thrive at high temperatures, and is almost always absent from the water during the summer months, or when the temperature of the water is above 55° F. Only once in the last four years has a growth of Synura been found in Boston water between May and October. The exception was in September, 1891, in Lake Cochituate, where there was a considerable growth at the mid-depth; but even there the temperature was below 55° F. There are, however, rare instances in which Synura has been found in hot weather, as for instance in Walden pond, Lynn, Mass., in August, 1891.

In September, 1891, Synura was present at the mid-depth of Lake Cochituate, where it imparted a slight taste to the water. Its distribution at this time was something peculiar. The growth was confined to the vicinity of the deep hole near the gate house, and, moreover, was found only in a stratum about 10 feet thick, about 55 feet below the surface. The temperature of this stratum was between 48° and 50° F. The layer of water immediately below the Synura had a decided cloudiness and contained considerable Crenothrix. These conditions prevailed for about a month, during which the Synura varied from 20 to 70 standard units per c.c. (One standard unit equals 400 square microns.) The

following table shows the state of things on September 28, 1891:

Depth in feet.	Color.	Temperature.	Crenothrix per c.c.	Synura per c.c.	Cloudiness.	Taste.
35	0.50	49°	0	0	0	0
40	0.55	48°	10	25	slight	slight
45	0.95	45°	156	0	distinct	0
50	2.40	44°	32	0	0	0

In January and February, 1892, Synura was again present in Lake Cochituate immediately under the ice. While the numbers were not large, the conditions for the production of oil were probably at their best, for the taste was strong. This taste and the Synura colonies themselves could be traced through Chestnut Hill reservoir into the service pipes, where in certain parts of the city the taste was quite strong, and complaints were made by the consumers. It is likely, however, that other infusoria than Synura helped in the production of this taste. That the taste was not due to the decay of the organisms in the pipes is shown by the fact that the bacteria at that time were quite low, the average of 14 tap samples being 61 per c. c.

The most extensive growth of Synura which has been found in Boston water occured in the ponds on Stony brook just above basin 3 in November and December, 1893. Both in Rice's and in Nichol's mill ponds the number of colonies frequently reached 200 per c.c. (equal to about 1,000 standard units.) These were gradually washed down into basin 3. At one time 2,000 standard units were found in the influent stream. Very few, however, reached the city taps, and no trouble was caused.

There is no question but that the Synura uvella is a very objectionable organism. Mr. F. F. Forbes,* Super-

* F. F. Forbes. The Relative Taste and Odor Imparted to Water by same Algæ and Infusoria. Jour. N. E. Water Works Association. Dec. 1891.

intendent of Brookline Water-Works, has stated that 10 colonies per c.c. will render a water unfit to drink.

From our experience it is certain that 10 colonies of Synura per c.c., if they are in the right condition, will cause a taste sure to be noticed by the consumers.

Typhoid Fever.

By ELMER LEE, A. M., M. D.
CHICAGO, ILLINOIS.

Recognition of the value of cleanliness represents the most practical discovery in treatment during the present generation, and, at the same time it constitutes one of the really great discoveries in the history of medicine. The application of the principles of cleanliness more nearly meets the requirements of a real advance in curative medicine, than all the other propositions known to the profession for the cure of disease.

The symptoms of Typhoid Fever are too well-known by all to need particular mention; the question of burning interest is what to do to be saved. The disease is produced by drinking contaminated water, and its seat of development is situated in the intestinal canal. There is a poison there which, if it could be removed before it had become absorbed into the blood, life, and even health would be spared. Allowed to remain, the poison is drawn into the circulation, and very soon the whole body feels the depressing effect. Even at this time, if those remaining poisonous juices and germs which are contained in the bowels were either neutralized by suitable remedies, or washed entirely away by a stream of flowing water, the disease would be checked, the patient spared, and health restored.

The Typhoid fever patient receives as food, whatever is simple, at regular intervals of four hours. Milk, simple, natural milk, is nourishment of the highest im-

portance. One egg every day, or every other day, is alternated with a small teacup of fresh pressed juice from broiled steak or mutton. The egg is pleasant to take and more nutritious, when whipped till it is light and then stirred with a small glass of milk. For a simple and nourishing artificial food, malted milk is always good.

The juices of fruits are delicious to the Typhoid fever patient, and are not to be dismissed on the supposition that they are injurious. It is always interesting to observe that, when the fever is broken, and convalescence is begining, water in copious draughts is no longer easy for the patient to take. When the usual glass of water is handed back half drained. it is an encouraging sign of beginning restoration. For wholesome drinking, fresh lake water which has passed through a Pasteur porcelain filter is entirely reliable.

The simplicity of the foregoing plan meets every requirement, and saves nearly every case, unless there is some complication. It is my belief that doing more than this is doing less, and less than this which is so simple, is not enough. The profession agrees that no kind of drug treatment is useful or curative in Typhoid fever, indeed, one of these days, in my opinion, the statment will be considered applicable to other, if not all, cases of diseases of the bowels.

The plan as proposed by me and practiced during a period of five years, consists, in review, of the following systematic management in Typhoid fever.

Water used internally as a douche for free irritation of the bowels, either simple or made soapy with pure liquid soap. Water as a drink, and as a remedy taken copiously and frequently, especially during the stage of fever. Water is indispensable, and should be given as often as is desirable and agreeable to the circumstance of the case. Frequent application of cool water to the

surface of the body during the entire illness.

Remedies: Peroxide of Hydrogen (Marchand's), or Glycozone, for the anticeptic effect of the oxygen which is set free in the stomach and intestines. But to be of real value, these remedies are to be taken in considerable quantity largely diluted in water, else, in my opinion, they are of little use. The capacity of the bowels is so great that a little of anything cannot spread over this enormous area to effect it beneficially. Cleanliness is the principle governing the use of Peroxide of Hydrogen (medicinal) and Glycozone.

For a remedy that sooths and brings on sleep at night, sulphate of Codeine is better than chloral, besides it is the safest and best.

For food, anything that is simple and in liquid form. Milk is always the best; milk and whipped eggs; pressed juice from broiled meat. The juice from fresh, ripe fruit. The nutrition taken should be at regular intervals (four hours), that sufficient time may be allowed for digestion.

Stimulants and drugs are injurious without exception, and better results are secured without their use. Typhoid fever, generally transmitted through the drinking water, is a preventable disease. Typhoid fever affects all classes, but if food and water were always pure, no class or age need contract Typhoid fever. Cleanliness everywhere and always is the means at hand which makes it possible to escape Typhoid fever and other diseases of the bowels. Internal cleanliness as well as external is a reasonable proposition of hope for the cure of the unhappy multitude of sick and discouraged humanity.

Centrifugation—to separate sediment, is coming into general use among European investigators. A number of instruments are in use for the purpose.

Microscopical Technique Applied to Histology.—VI.

[FROM THE FRENCH OF RENE BONEVAL.]

Continued from page 248.

Permanent Preparations of Blood.—. . . .Spread a blood drop as thin as possible on a slide. Pass to and fro over an alcohol flame until dry. The corpuscles are thus fastened to the glass in their natural form and size. . . .

Frog's blood fixed by heat may be stained by eosine and by methyl green. Stain for 3 minutes with a few drops of the following: eosine, 2; water, 50; alcohol, 50. Dissolve the eosine in the alcohol and add the water. Wash gently, and stain for 2 minutes with an aqueous solution of methyl green. Wash, dry and mount in balsam, after clearing by oil of bergamot. The globules are red, the nuclei and leucocytes green. . . .

To preserve the red corpuscles in a tissue to be sectioned use, as a fixative, picric acid or 2 per cent ammonia bichromate. Complete the hardening by alcohol, imbed in gum; the corpuscles will not be changed as they would be if the tissue was fixed by alcohol alone. Stain the sections by hæmatoxylin and eosine; mount in balsam. . . .

FIBRIN.

. . . .A capillary tube with excedingly thin walls, filled by being thrust directly into a frog's artery, is placed on the microscope stage and examined with a high power. "At first the red globules fill the tube. In a few minutes coagulation takes place, and the cylindrical mass imprisoning the globules is seen to be separated from the glass by a transparent space which contains not a single corpuscle. Then the white corpuscles begin to leave the coagulum and to float in the serum. Seeing the activity of these amœboid movements, one would be tempted to attribute them to the escape of these anatomical elements from the clot, but this is an illusion as will

be shown hereafter. In a little while (usually in 45 minutes from the beginning of the observation), the red corpuscles begin to present the same phenomenon. They escape in such numbers from the sharply defined edges of the clot, that the liquid soon becomes so filled with them that further microscopical examination is impossible. If the tube be now removed from the stage and placed in a vertical position, in a short time the corpuscles will fall to the bottom, leaving a clear space above filled with serum. We might be disposed to consider this the dissolution of the coagulum, but the appearance is deceptive, for if the contents of the tube be forced out by the breath, the clot will be seen in the form of a slender cord of fibrin floating in the liquid. The clot is not dissolved, but the contraction of the net work of fibrin makes the meshes too small to contain the serum and the corpuscles at first imprisoned in the coagulum."

To Study the Net Work of Fibrin.—. . . .A drop of a frog's or of a mammal's blood is placed on a slide and left to coagulate in a moist chamber. When the blood is so firm that it may be inverted without displacement, immerse it in a vessel of filtered water. The blood will soon be decolored; remove the slide gently, and stain with the iodine solution or with eosine. Cover and examine in the staining solution. Iodine preparations are not permanent; to preserve the eosine specimen run under the cover a drop of salted glycerine colored with a little eosine. Salted glycerine, which will preserve aniline stain for a long time, is made as follows: Shake together 40 grms. of glycerine with an excess of common salt. Let it settle, then decant. . . .

THE HEART.

To study the cardiac muscle cells.—. . . .Dissociate these in a fragment from a frog's auricle by the aid of the 40 per cent caustic potash solution. These preparations

are excellent, but not permanent.... To observe the cells of Purkinji select a sheep's heart.... By superficial incisions circumscribe a square of endocardium, and remove it with a razor, taking great care not to trench to the slightest degree upon the myocardium. Divide the piece in two; carefully spread one portion face upward on a slide; if few fibres of the myocardium remain, stain with picro-carmine and mount in glycerine, but if they are in great numbers and mask the fibres of Purkinji, remove them by brush and forceps before proceeding further. The second portion being spread on the slide, add a drop of 40 per cent potash solution, and cover; to dissociate the cells forming the fibres of Purkinji press on the cover with a needle.

To make tranvers sections of the fibres.—Picric acid and ammonia bichromate answer very well to fix the tissue. Harden by alcohol, imbed in gum. Sections made after the action of the bichromate, should be stained by hæmatoxylin and eosine, and mounted in balsam; after picric acid, stain in picro-carmine and mount in formic acid glycerine.

The endocardium and the valves.—The former may be well studied in sections made perpendicularly to the internal face of the heart. Fix by picric acid, harden in alcohol, imbed in gum; stain by picro-carmine, and mount in acid glycerine.

To see the endocardial endothelium select the heart of a rat or of a rabbit. Into a vein inject a solution of silver nitrate 1 to 300; until the liquid returns through the arteries. Wash the internal surface of the heart with the silver salt, open it in a vessel full of distilled water and expose to direct sun light. With a razor, cutting paralel with the surface, remove a fragment of the endocardium and spread it on a slide with the inner face upward. Stain with alum carmine, wash, dehydrate by absolute alcohol, clear by clove oil and mount in balsam.

The endothelium of the valves may be studied in the same way. To see the arrangement of the connective layers forming the frame work of the latter, proceed thus : Pin a valve to a cork and immerse in picric acid. In 2 hours remove the pins and complete the fixing by leaving it in the liquid for 6 hours. Harden in alcohol, imbed in gum. Cut longitudinal and transverse sections; stain by picro-carmine, examine in acid glycerine.

ARTERIES.

. . . .*Sections.*—After removing the greater portion of the cellular tissue about the aorta, open it lengthwise by scissors. Cut out a little rectangular piece so that its long sides shall be perpendicular to the lumen of the vessel. . . . If we desire sections parallel with the axis of the artery we cut parallel with the small side of the piece ; if perpendicular sections are wanted, section parallel with the larger side. This apparently puerile suggestion is of great importance. . . . The artery should be treated as were the valves. (Fixation by picric acid after extending on cork, hardening in alcohol, imbedding in gum). Sections parallel with the axis are most instructive. Deeply stain with picro-carmine, wash, mount in water, and allow the following liquid to run under : Glycerine, 50; formic acid, 1; saturated solution of picric acid, 50.

To dissociate the muscle cells from the middle layer, macerate a piece of aorta for 24 hours in the $\frac{1}{3}$ alcohol; with curved scissors remove a bit of the middle coat and spread on a slide with needles. Stain with picro-carmine, mount in that liquid after partial drying; replace the picro-carmine by a drop of glycerine.

The elastic plates may be easily demonstrated by dissociating a shred of aorta after macerating in a 1 per cent aqueous solution of tartaric acid.

The endothelium of the internal coat may be shown by

removing a portion of the thoracic aorta from a recently killed rat, slitting it open, placing it in a saucer of silver nitrate solution (1 to 300), and exposing to direct sun light. It is necessary to keep the tissue in motion during the entire exposure. Wash, mount in balsam, the internal face of the artery upward.

The smaller arteries may be studied in a transparent membrane. Fix the mesentery or the omentum in 2 per cent bichromate, wash, spread on a slide, stain with hæmatoxylin, mount in balsam. The internal elastic membrane will be visible in a great many organs when the vessels have been sectioned transversely. . . .

VEINS.

On account of the differences in the structure of veins, many preparations must be made from different vessels. Employ the same processes as for the arteries.

CAPILLARIES.

The structure and arrangement of the capillary net work can be well studied only by injections. We will examine the instruments used to make an injection, the injection masses and methods.. . . .

Instruments.—A good syringe is the simplest and most useful. Among the numerous models choose one holding about 250 grammes. The canulas should be fastened in place by friction; those that screw on are detestable. It is well to have a supply of cylindrical canulas with a neck at the free end for fastening the ligature. . . .

Injection material.—From the great number of these we select the two which are most frequently used, the Prussian blue gelatine and the carmine; we advise beginners to use the former.

Soluble Prussian blue mass.—Soluble Prussian blue may be purchased of the dealers, or prepared as follows: Make these two solutions (1)—Distilled water, 1000 grms.; sulphate of iron, 50 grms. (2)—Distilled water,

1000 grms. ; ferro-cyanide of potassium, 100 grms. Mix solutions (1) and (2). They form a precipitate of insoluble blue. Pour on a filter and wash until the liquid runs blue below the filter. Dry, scrape off the precipitate and keep for use.

For an injection, make a saturated aqueous solution .... Soak 10 grammes of gelatine in distilled water for 20 minutes, then remove it from the water, and melt it in a water-bath. In a water-bath heat 200 grammes of the blue solution, and when this and the gelatine are at the same temperature, pour the blue into the gelatine, and stir with a glass rod. A precipitate is formed, but dissolved as the heat increases. The solution is perfect when the glass rod shows no blue grains on its surface. Filter through flannel and keep the temperature at 40° C. until ready to use. If any remains after the injection is made, a little thymol should be added, otherwise the gelatine will liquefy in a few days and the material be destroyed.

(*To be continued*).

Notes on the Habitat of Diatoms.

By R. D. NEVINS,

BLAINE, WASH.

The collector may think himself fortunate if he finds any locality or discovers any habit of growth of these lovely things which will enable him to secure them specifically separate or nearly so and already cleaned by processes of nature.

I will give some notes of what have been to me lucky "finds," hoping to provoke from others reports of similar good fortune.

Zostera marina and another species which we have on Puget sound, Z. oregana, have been to me treasures in this respect. The plant is found everywhere floating its

long ribbon-like fronds upon the surface at low tide, and always in the tangle of sea-weed which is rolled by the tide and wind on the shores.

Arachnoidiscus Ehr. often studs this frond thickly. It may be felt by the hand as the frond passes through the fingers as one almost involuntarily reaches for it as he passes over the ground which it frequents and it may be seen glittering on the frond which it often encrusts when the sun has dried the tangle on the beach.

Isthmia nervosa loves the same habitat, and it laces the surface and fringes the edges of the frond with its zigzag chains.

This plant is also often gray with encrusted Coconeis scutellum, and others of the same genus or is frosted with Nitschia or Synedra.

On other sea weeds I have found Triceratium arcticum in patches, woven and interlaced, chain on chain.

An inexhaustible and clean find of Atthya decora rewarded my curiosity in noticing that the surf along the Pacific shore was fairly yellow with some floating substance. This I have seen twice for a week at a time in midsummer.

The bronzed film that shines upon the surface of the mud flats is full of interest because it seems composed of diatoms in mass, pleurosigma mostly with occasional Surirella gemma, all far more beautiful when found than they can be after being cleaned for mounting. The writer would be glad to know how to separate and clean them in any quantity proportionate to their number as found.

Notes of personal observation might be drawn out to great length and if from dilligent and careful collectors would be full of interest. I have in these here given only spoken of some of my most lucky finds. I should be glad if others would do the same.

Smegma Bacillus.

By PHILIP JAISOHN, M. D.
WASHINGTON, D. C.

Of late years the practitioners can make a positive diagnosis of tuberculosis by examining microscopically the discharges of the suspected patient such as sputum, pus or urine as the case may be. The main diagnostic point of tubercle bacillus is that it retains the carbol-fuchsin stain even after treating it with an acid solution. On account of this peculiarity we can readily recognize it, as all other micro-organisms lose the red color as soon as an acid solution is applied.

There are numerous methods of staining tubercle bacillus, but the easiest and surest are those of Ziehl-Neelsen and Frankel-Gabbett. However, every method has one common object in view, that is to either decolorize or counter stain all other micro-organisms so that the red stained tubercle bacillus will stand out conspicuously in the field. Now there is another bacillus which has the same peculiarly, namely, the smegma bacillus. This organism had undoubtedly misled many a microscopist in the past and will do so in the future, if one's attention is not called to it. Prof. Welch of Johns Hopkins Hospital first called my attention to the fact, and recently I made some study on the subject. I obtained a large quantity of smegma from some negro patients who have no tubercular history, and made a number of cover slip preparations from this material, and stained them just like staining for Tubercle bacillus, (Modification of Ziehl-Neelsen's method). The field under the microscope is identical with that of tubercle bacillus, except the red-stained bacilli in the blue back ground are somewhat broader and more curved than tubercle bacilli. Although if I had not known that it was smegma I would not have noticed this difference. I

mixed some of the smegma with 500 c.c. of healthy urine, and let it stand for 24 hours, then examined the sediment in a like manner. The result is the same as pure smegma, except the bacilli are less curved and considerably shrunken so they look more like tubercle bacilli than before mixing with the urine. It seems to me very important to know this fact, especially those who use a microscope must remember that there is an organism other than tubercle bacillus which resists the action of an acid and retains its fuchsin stain. This is specially important in examining urinary sediments for tubercle bacillus. I have already stated that the size and shape of smegma bacillus are different from that of tubercle bacillus to the eyes of those who are familiar with the morphology of the latter, but in the case of doubt stain it by the Gram's method as the former takes the stain very promptly, while the latter takes it very feebly and the protoplasm of the rods is contracted into a series of spherical, stained bodies resembling a number of micrococci.

The Meeting of the American Microscopical Society at Brooklyn, N. Y.

By W. H. SEAMAN, SECRETARY,
WASHINGTON, D. C.

The Seventeenth Annual Meeting of the American Microscopical Society was held at Brooklyn on August 13, 14, and 15 in the Polytechnic Institute. In common with some of the other scientific societies which met at the same time and place the first day was not very successful. The President did not arrive till the afternoon and it seemed as if very few of the local microscopists took any interest in the meetings.

The second and third days were very satisfactory, the attendance being quite as large as last year and the character of the papers excellent.

The address of the President, Dr. Lester Curtis, of Chicago, was delivered on Tuesday evening, his subject being "Microscopical Reminiscences."

The following new members were elected: Dr. John L. Gilbert, Topeka, Kansas; Miss Eva H. Field, Des Moines, Iowa; Miss Agnes M. Claypole, Akron, Ohio; Benjamin F. Kingsbury, A. B., M. S., Defiance, Ohio; Dr. Charles Franklin Craig, 21 Balmforth avenue, Danbury, Conn.; Dr. Walter W. Alleger, Pension Office, Washington, D. C.; Joseph D. Mallonee, 84 Fargo avenue, Buffalo, N. Y.; Karl McKay Wiegand, Ithaca, N. Y.; Dr. Albert H. Brundage, 1153 Gates avenue, Brooklyn, N. Y.; Dr. Jos. H. Hunt, 1085 Bedford avenue, Brooklyn, N. Y.; Dr. Bushrod W. James, N. E. corner Green and 18th streets, Philadelphia, Pa.

Seven papers and accompanying slides and photos were presented for prizes, and the Executive Committee, after careful consideration, awarded the prizes as follows:

To Karl McKay Wiegand, of Ithaca, N. Y., for his paper, "The structure of the fruit in Ranunculaceæ," there being no competition in this class, the second prize of thirty dollars, for original research in botany; to Benjamin F. Kingsbury, of Defiance, Ohio, for his paper, "The histological structure of the Enteron of *Necturus maculatus*," the first prize for original research in Zoology of thirty dollars; to Agnes M. Claypole, of Akron, Ohio, for the second best in Zoology, on "The Enteron of the Cayuga Lake Lamprey," the second prize in Zoology of thirty dollars; to Dr. Charles M. Krauss for set of photomicrographs, there being no competition in this class, the second prize of fifteen dollars.

In addition to the above, the following papers were presented:

A contribution to the study of diseases of the lower animals, by Miss Eva M. Field, of Des Moines,

Iowa; a contribution to the study of the Burrill corn disease, by Dr. V. A. Moore, Washington, D. C.; a new method for securing paraffin sections to the slide by Miss Agnes M. Claypole, Akron, Ohio; a marker for indicating the position of objects in microscopical preparations, by Prof. S. H. Gage, Ithaca, N. Y.; formalin, by Dr. J. Melvin Lamb, Washington, D. C.; simplification of laboratory methods, by Dr. Wm. C. Krauss, Buffalo, N. Y.; limitation of tuberculosis, by Dr. W. W. Alleger, Washington, D. C.; a study of the muscular tunic in the large and small intestine of man in the vicinity of the Cæcum, by Dr. R. O. Moody, New Haven, Conn.; a study of the microscopic phenomena of inflammation, with special reference to the Diapedesis of the white-blood corpuscle, by Dr. Charles F. Craig, Danbury, Conn.; a third study of the blood hæmatoblasts and plaquelettes, by Dr. M. L. Holbrook, New York city.

The following officers were elected for the next year:

President, Prof. Simon Henry Gage, of Ithaca, N. Y.; Vice-presidents, Dr. Veranus A. Moore, of Washington, D. C., and Henry G. Hanks, of San Francisco, Cal.; Treasurer, Magnus Pflaum, of Pitsburgh, Pa.; as members of the Executive Committee, Dr. Robert O. Moody, of New Haven, Conn.; Charles S. Schultz, of Hoboken, N. J., and Prof. Henry B. Ward, of Lincoln, Neb. The term of the Secretary has not yet expired.

Some new apparatus was exhibited by Messrs. Woolman and Drescher, and on Tuesday afternoon a very interesting visit was made to the Hoagland Laboratory.

The Executive Committee decided that it was inexpedient to hold an exhibition and working session this year. These portions of the exercises of the annual meeting depend very much upon the local microscopists of the place where the meetings are held for their success, and it did not appear that the conditions at Brooklyn were such as to encourage an attempt. In regard to

these adjuncts it would be very desirable that some members of the Society should take them in charge and make them as successful as they have been on past occasions. There are many members who think that they are among the chief attractions of our meetings, and that if fully developed they would secure a larger attendance. It is manifestly impossible for the Secretary to assume charge of these in addition to his other duties. Cannot some method be suggested by which something of the kind can be carried out, if not on so large a scale as formerly, yet so as to attract those to whom demonstrations are particularly interesting? At Madison the talk by Prof. W. S. Miller really occupied one afternoon, and was a working session for the work of one man. Could not some member at each session, even if no elaborate preparation was made or public invitations extended, give his individual demonstration as to how he works in his particular line.

It is evident that the prizes have been a stimulant in the matter of high-class papers, and it would be very desirable if those for the papers should be continued. It was suggested informally that the Spencer Tolles fund should be raised to a thousand dollars by subscriptions of one hundred dollars each from several cities, and the interest used as a premium fund. It ought not to be a very difficult undertaking to do this in ordinary times, but the exceptional financial conditions now prevailing are adverse. Nevertheless, the sum required is very small, and we believe that a little effort on the part of our members would effect the result. Some such action as this would do more to render the Society permanent and successful than anything that can now be done.

It should be remembered that the amount offered in no sense represents anything like payment for the work done, that the honor is more than the money; but in at least one case the award aided the recipient in securing

a permanent and honorable engagement, and if an endowment could be raised by which the offer of premiums became a regular feature it would be a great stimulus to advanced biologic work in this country.

It is anticipated that the next meeting of the American Association for the Advancement of Science will be held at San Francisco, California, next summer, and under the present arrangement our Society may go there. We shall, of course, have the benefit of all the special rates that may be accorded, but to most of our members it will appear a long journey. A very cordial invitation in writing was received from the citizens of Chattanooga, Tenn., through Dr. James E. Reeves, to meet there next summer, and it will become the duty of the Executive Committee to consider the matter in due season.

Preparing Teeth for Microscopic Study.

BY W. E. CHRISTENSEN, D.D.S.

When the tooth has been extracted, it must immediately be brought into some conserving fluid—as alcohol or watery solution of bichloride of mercury—to prevent the pulp from shrinking. Also, in all further manipulations as cutting, sawing, or grinding the sections, careful attention must be given that the tooth or section is at no moment without an excess of moisture. If teeth of rabbits or dogs are used, it must be remembered that the tissues of the pulp stick more to the wall of the pulp chamber than in human teeth. This is important at the first opening of the tooth. If human teeth are taken from dead bodies, it must not be more than six hours after death.

In order to enable the preparing drugs to enter the pulp, it is necessary to open the tooth and partially expose the pulp in such a manner that the pulp is not hurt. For this purpose we make a section with a sharp, fine

saw on both sides of the tooth about midway of the crown, and cut off the part with a pair of strong excising or splitting forceps. The end of the pulp will then be exposed without being hurt. At the apex of the root a short piece is cut off in a similar way. For fixation of the tissues, the tooth is placed in a saturated solution of bichloride of mercury in water for about eight to twelve hours, after which it is washed in running water for two hours.

To harden the tissues of the pulp, the tooth is then placed in alcohol of low degree, the strength of which must be gradually increased from 30 to 50 per cent., and from 50 to 70 per cent., leaving the tooth from eight to twelve hours in each of the three stages. From the bichloride of mercury there will be left a black precipitate on the specimens which can now be removed by placing them for about twelve hours in 90 per cent. alcohol, to which 1½ to 2 per cent. tincture of iodine has been added. The iodine is removed by placing the specimens in pure alcohol until they recover their former white appearance. After the process of hardening in alcohol, the teeth can be cut in two or three sections by means of a fine, sharp scroll-saw. The next step is to stain the specimens. There are a great number of dyes used for staining preparations for microscopic studies. Dr. Weil, in the description of his method, mentions a great many, but he says he has obtained the best results with "aqueous borax-carmine," prepared in the following manner :—

Eight grammes of borax is powdered in a mortar together with 2 grammes of carmine, and then mixed with 130 grammes of aq. dist. After twenty-four hours the solution must be well filtered, and is then ready for use.

The sections, when taken from alcohol, are washed in running water for about fifteen minutes, and then transmitted to the dye for twelve to twenty. hours, in which

time they will become intensely colored. The excess of dye is removed by placing the sections in 70 per cent. alcohol, to which 1 gramme of hydrochloric acid has been added to each 100 C.cm. for as long as they have been in the dye. It is then well to place them in 90 per cent. alcohol for about twelve hours, and afterwards in oil of cloves for one or two hours. This last procedure makes the specimens more transparent.

We have now come to the treatment with Canada balsam. This must in the meantime have been prepared in the following manner: A quantity of Canada balsam, as obtained from the druggist, is placed in an evaporating dish, and kept over boiling water until the balsam, by cooling, becomes solid and brittle like glass. This takes from twenty to thirty hours, according to the quality of the balsam.

When the sections are taken from the oil, they must be cleaned with pure xylol, and then quickly immersed in chloroform, to remain twenty-four hours. Then sufficient of the hardened Canada balsam is added to the chloroform to give a weak solution, and after another twenty-four hours the solution must be saturated with Canada balsam; but the solution containing the specimens is then again put into an evaporating dish, and placed over hot water until the balsam once more becomes solid. As the chloroform boils at a low temperature (60-61° C.), the heat from the water must, to begin with, not be higher than about 60° C.; later it can be increased, but altogether slowness of the solidifying process is in favor of the results. Consequently, the procedure takes from twenty to thirty hours.

When the balsam, by cooling, becomes solid and brittle, the specimens can be cut out with a small, sharp chisel, and with a fine, sharp scroll-saw be cut into sections suitable for grinding. The grinding is done as when small sections of bone are ground for microscopic

study, always under application of excess of water. The pressure must be very gentle, especially when the specimen gets thin; the result now depends somewhat upon the skilfulness of the operator. When the section is thin enough, which must be controlled under the microscope, it must be carefully cleaned in distilled water, and finally mounted in Canada Balsam.—*Dental Cosmos.*

EDITORIAL.

Oliver Wendall Holmes, a Microscopists.—Forty-one years ago, Dr. Holmes who was 85 years old on August 29, 1894, taught Dr. E. Cutter how to use the microscope with direct illumination. He had an arrangement of his own,—a six-inch black disc fastened to the tube and graduated so that turning the disc would act as a fine adjustment. Dr. Cutter says that Dr. Holmes worked a good deal with the microscope in those days and that the intellectual drill derived therefrom may have been used in literature. Is not the technical use of the microscope in college as good a discipline as the study of Greek? Surely the cyclops of the Odessey would be better understood by one who has studied a living cyclops taken from a hydrant and shown under the microscope.

Centrifugation—to separate sediment, is coming into general use among European investigators. A number of instruments are in use for the purpose.

Photo Micrographs.—At the British Medical Convention, Mr. Andrew Pringle made the following exhibits and explanations.

At schools of medicine and the like, where a large number of students are engaged in studies which are only made possible by the use of the microscope, it has been customary to cut a large number of sections from a piece of tissue, and to examine such sections, each under a different microscope, for the microscope is an instrument which permits of only one observer at a time. Now, no two sections can be identical, and more often than not, critical points, upon a right appreciation of which the subject-matter of a lecture may altogether depend, are absent i ı

some; but, if the demonstrator be provided with an optical lantern, and he is able to exhibit the photograph of a chosen section of the tissue under consideration, he can point out its salient features to all the students at the same moment, and no one can complain that he is not as well off as his neighbor. Mr. Pringle then proceeded to give evidence of the advantages which he claimed for lantern demonstration by exhibiting a large number of photographs on the screen. First he showed some photographic preparations, which were exceptionally fine, among which were a slide of "voluntary muscle" by himself, an example of ossifying cartilage (Klein), a complete section of the human eye, showing very beautifully the cornea, crystalline lens, the retina, and the optic nerve, and a fine sample by Bousfield of Cortis' organ of the inner ear.

Next came some very beautifully executed photo-micrographs of bacteria, including a "plate culture" of Proteus (Klein); two examples, showing bacteria in the dentile tubules of decaying teeth (Sewill), and a splendid example of anthrax bacilli in mesentery, by Pasteur.

The consideration of preparations of great rarity came next, and as an example of these were shown that happily rare organism known as *Filaria sanguinis hominis*, a parasitic, worm-like creature, which, as its name implies, finds its habitat in human blood, but more particularly in the blood of negroes. Three slides referring to this interesting organism came under review by the lecturer, showing—(1) its ordinary appearance; (2) the sheath of the parasite; and (3) the number of parasites in the restricted field of the microscope; and the comparative size of *F. nocturna* (so named because it is only found in the night time) and *F. perstans*.

Then came a few words about objects which exhibited unusual difficulties in preparation, and as an example of these the lecturer exhibited a photograph of *Cholera Bacilli*, with their flagella plainly visible. These flagella are most difficult to see, even to a trained microscopist, and it might easily have been asserted that a man had fancied he had seen them, and that they did not really exist; but here was the photographic record which cleared up the doubt conclusively. Another example of the *Bacillus termo*, also showing flagella, was thrown upon the screen, and the lecturer explained that in this case the hairlike flagella had

been carefully measured by Dr. Dallinger, who had found that they had a width of only the 1-200,000 of an inch. It was quite a triumph to reproduce these microscopic objects photographically.

The lecturer next dealt with examples which, by speedy and convincing comparison, would serve to set at rest questions in which opinion was divided. He showed two preparations of nerve cells, the one being properly "fixed," and the other being faulty in that respect, giving rise to appearances which might lead a student into error. Examples were also shown of certain newly discovered bodies which were found in cancer, the real and the spurious being readily distinguished by the comparative method.

The next slides shown were of very great beauty. They were "culture plates," which had been exposed for five minutes, the one in the comparatively pure air of Wadsworth Common, and the other in a wide street. The first-named showed only a few traces of bacteria, but the second was crowded with thick colonies of them. The advantage of at once obtaining photographic records of preparations subject to such quick growth and other changes as these culture plates are, was fully pointed out by the lecturer. In a demonstration of some particular points where only exceptional microscopic preparations are available, the lecturer showed as examples (1) the position of the bacillus of leprosy in regard to cells and nuclei, and (2) the position of the organisms in certain vessels in a rare form of skin disease. In this last demonstration three slides were exhibited of the same section, each under a different amount of magnification. In the first, taken with a low-power objective, the organisms were hardly visible; in the second they were plainly seen; and in the third, where the magnification was 1000 diameters, they came forth in all their hideous detail. Some specimens of lantern slides, colored by the Lumiere method, elicited the fact that, although the original preparation was well imitated so far as the staining was concerned, the advantage gained was more than counterbalanced by loss of crispness, and the lecturer expressed a doubt whether, under such conditions, there was any gain at all.

In order to convey an idea to his audience of the actual magnitude of some of these microscopic objects, the lecturer threw

upon the screen photographs of yeast cells, in conjunction with a network of squares, the sides of which measured only 1-3750 of an inch. In the case of some other organisms, squares measuring 1-7500 of an inch were employed.

MICROSCOPICAL APPARATUS.

A Convenient Sub-stage Condenser.—A periscopic eye piece set in the sub-stage with an adapter will afford a very good sub-stage condenser.

Polarizers are usually fitted with an adapter which may be removed and used with eye-piece in the sub-stage. The condenser certainly improves the performance of the microscope, it renders the light more pleasant to the eye and better defines the objects. Often, things hardly visible without the condenser, for example, bacilli tuberculosis, become distinct when it is employed. With low powers the condenser should be moved down away from the objective and with high powers brought up close to them. No one who has ever used a condenser would be without one. The light is also much improved by using a microscopic lamp with a blue glass inserted in the bull's eye.—L. C. W.

MICROSCOPICAL MANIPULATION.

New Method of Staining Micro-Organisms in the Blood.—A comunication on this subject was made by M. H. Vincent at the last meeting of the Societe de Biologie.

The process is applicable to every variety of micro-organism occurring in the blood, but is particularly serviceable in microscopical examinations in bacteria, which do not take coloration by the method of Gram, and of various parasites, such as hematozoa of paludism.

The process is based on the following principle, viz., coloring matters fix themselves not to the protoplasm, but to the hemogloben itself. If, therefore, the latter color-taking constituent be made artificially to disappear and the coloring agent be then brought to bear, the blood globules which mask the bacteria become invisible, and the microbes alone remain colored and stand out under the microscope with great clearness.

Among the different solvents, tried M. Vincent has selected the following liquid, which does not alter the form of the globules, and leaves no deposit and no striæ:

Aqueous solution carbolic acid, 5 p. c........	6 centim. cubes		
Water saturated with Na. Cl..	30	"	"
Glycerine...	30	"	"

Filter.

The blood spread in a thin layer (or in a thick layer when microbes rare in number are being sought) is slowly dried, either at ordinary temperature or a feeble heat. It is then treated with the fluid above described, which dissolves entirely the hemoglobin. At the end of half a minute to two minutes the fluid is drained off, the blood washed with distilled water, and coloration is effected with carbolized methylene blue, with the addition of from 1 to 20 per cent of aqueous solution of methyl violet.—*Paris Cor. Med. Press and Circular.*

Silverin, a New Polishing Material.—This consists, according to the *Pharmaceutische Centralhalle*, of 30 parts of precipitated chalk, 30 parts of ammonia, 45 parts of alcohol, and 200 parts of water. Shake before using.

Seeing Bacillus Tuberculosis with a Low Power.—If properly stained this object can be seen nicely with a one-quarter inch objective. All that an amateur would care to see can be thus exhibited. If stained red on a blue ground it will appear plainly and beautifully.

MEDICAL MICROSCOPY.

The Bacteria of Rheumatism.—Professor Max Schuler is said to have discovered, in the joints of persons attacked with chronic articular rheumatism, bacteria, which are always identical in like cases. These bacilli are short and thick, having at each end bright grains which aniline colors make still more evident. The discoverer has been able to cultivate these bacteria in bouillon, or gelatine, or on a piece of potato. Their culture requires a temperature of at least 25 degrees, and darkness is indispensable.

Cancer.—There is a growing opinion that cancer is ascribable to the action of a gregarine. These singular corticate infusoria will be familiar to students who have brought a micro-

scope to bear upon the tissues of an earthworm; indeed, it would be hard to find a specimen not infested by some samples of this group of Protozoa. In the invertebrata, however, they do not appear to be fatal to life, or, indeed, to cause any very great inconvenience, and these recent researches of Drs. Ruffer and Walker invest the group with a sinister human interest it has not hitherto possessed.

BACTERIOLOGY.

Bacteriology of the Embryo.—Maffucci gives his results of one thousand bacteriological experiments on the chick embryo and of one hundred and fifty on the fœtus of the rabbit: he draws the general conclusion that whilst the embryo lives it does not permit the development of pathogenic germs in its tissues save in exceptional circumstances, but may destroy, attenuate, or store up such germs for later deveolyment in its extra-ovular life. He states that in some cases a non-pathogenic virus of the adult fowl may be pathogenic as regards its embryo. These conclusions are founded more upon experiments on the chick than on the fœtal rabbit. The doctor gives the results of his experiments on the passage of tubercle bacilli through the placenta from the mother, and on paternal infection of the ovum with tuberculosis.—*Tertatologia*, April, 1894.

DIATOMS.

Staining.—A few years ago I tried some experiments in staining diatoms, hoping to thus secure a more pronounced relief of the delicate markings and a more perfect microscopic resolution of their structure. While in many instances the effects were fine, and the mounts reasonably permanent, I did not push the work to complete success. During the current summer I have employed a great deal of time in carrying forward this work, and with results even more surprising than my former experience warranted me in anticipating. The strength of image of the most delicate forms, and the exquisitely fine beading upon those diatoms which have been heretofore most difficult of resolution, have been wonderfully increased, making the study of their structure a matter of comparative ease.

The colors which I have used with the greatest success are

black, blue-black, green, red and brown. The stainings in black and brown are particularly good for study by lamp-light, and all of the colors named above give surprisingly strong images in photomicrography. I would suggest the importance of workers in microscopy carrying on this line of study, as the process of staining, renders possible much that was very difficult or absolutely impossible with the old method of treatment.—WILLIAM LIGHTON, *Omaha, Nebr.*

MICROSCOPICAL NOTES.

Microscopicol Praxis.—This is the name of Dr. Stokes' new book, just issued. It will be of much assistance to the novice but he who would go extensively into such matters will take up Carpenter or Gage after mastering the outlines here presented, and some may be induced to do so by the pleasing introduction here acquired.

To Keep Metallic Objects From Rusting.—Mix 11 parts of white wax and 2 parts of suint and dissolve in spirit of turpentine. With a soft sponge or other similar object apply the solution to the surface to be protected. Objects so protected will remain free from rust indefinitely.

NECROLOGY.

Ezra Hollis Griffith, A. M.—On August 18th Professor E. H. Griffith died at his home in Chicago, at the age of 56. He was born in Oneonta, Otsego County, N. Y., enjoyed a liberal education, and throughout his life was interested in literary and scientific pursuits.

In early life, while a teacher, Professor Griffith became interested in the use of the microscope. The desire for a portable instrument led to his invention of the Griffith microscope, a very noted and popular instrument among specialists. He was largely instrumental in organizing the American Microscopical Society, and for several years had charge of what is known as "The Working Session." In recognition of his services he was made a member of the Royal Society of Microscopists of London.

Being possessed of generous impulses he was easily touched

with the sufferings of others. The needy found in him a friend and the discouraged a helper.

MICROSCOPICAL SOCIETIES.

Quekett Microscopical Club.

At the last meeting Mr. C. L. Curties showed a new instantaneous photomicrographic apparatus, and described the method of using it. Some very fine pictures of pond-life, fresh human blood, etc., taken by this apparatus were handed round for inspection, and one group of *Lophopus* fully extended, surrounded by *Vorticellæ*, was particularly admirable and life like. The Chairman thought this apparatus would be especially valuable for obtaining representations of quickly moving organisms, which were almost impossible to draw in a natural way because of their rapid evolutions, and they might get composite pictures which would throw some light on this difficult subject of locomotion in minute animals, such as had been done by Muybridge and others with the horse, for instance. Mr. G. Western read some interesting notes on foreign rotifers which had since been found in Britain, amongst them being *Notholca heptodon*, *Bipalpus vesiculosus*, *Chromogaster testudo*, *Æcistes mucicola* and *Æ. socialis*, *Brachionus dorcas*, and others, which were accompanied by beautifull drawings by Mr. D. Nuttall.

Mr. Western pointed out the uncertainty and variability of many characters relied upon for specific, and in some cases for generic, value, such as the presence or absence of setæ, antennal appendages, or even of the eyes. Mr. Michael said, with regard to the eye, he had frequently found the same peculiarity among the Hydrachnea or water-mites; in the same gathering would perhaps be met with specimens otherwise identical, some with and some without eyes, or the eye present on one side only. The pigment greatly varied in amount, or was entirely wanting, but without sections it was difficult to say whether that was the case with the true nervous part of the visual organ, which from its transparency was easily overlooked in merely surface views.

Washington, D. C.

September 8, 1894.—The regular monthly meetings of the Society were resumed on Tuesday evening at No. 714 13th Street. N. W. On this occasion some interesting information relative

to the Hoagland Laboratory, Brooklyn, N. Y., was given by Dr. Seaman.

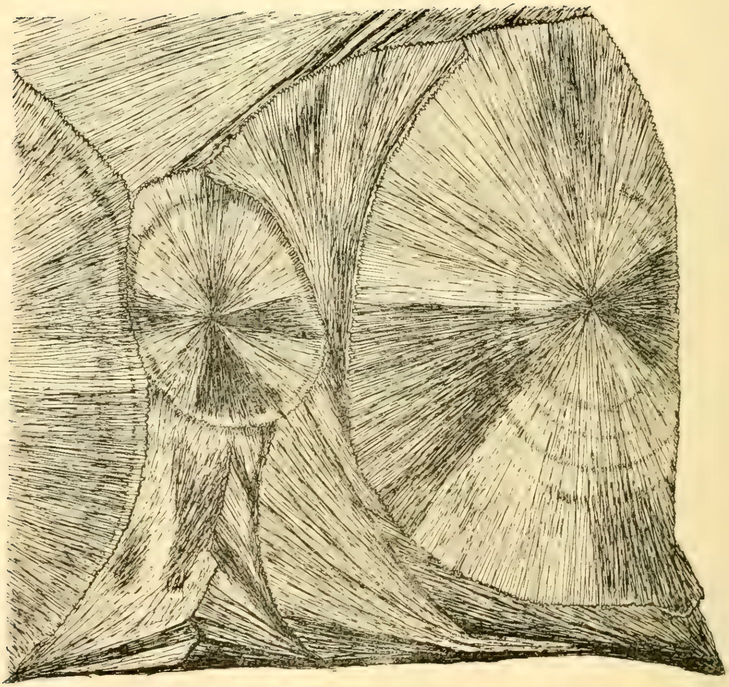
NEW PUBLICATIONS.

Mikrophotographie und die Projection. By Dr. R. Neuhaus. Halle a. S.: Wilhelm Knapp, 1894. Pp. 58; 5 illustrations. Price 40 cents.

This book treats microphotography in such a popular way that even a beginner, by following the rules laid down, will soon be enabled to produce satisfactory pictures. The second part of the book gives a description of how to project prepared microscopic objects on a screen.

Twelve Edible Mushrooms of the United States, with Directions for their Identification and their Preparation as Food. By Dr. Thomas Taylor, Chief of the Division of Microscopy of the Department of Agriculture.

The importance of the subject need not be insisted upon to the few who know how immensely superior the fresh American mushrooms are to those imported from Europe—as Mr. William Falconer of Glen Cove, N. Y., puts it, in a summary contained in Dr. Taylor's pamphlet of a paper read before the Massachusetts Horticultural Society for February. "Many persons who have used the tasteless, indigestible, putty-balls from imported cans will repudiate the foreign article and accept no other than the wholesome, toothsome and juicy domestic product." In this statement Mr. Falconer has reference to the time when, as he foresees, the production will have been so increased as to reduce the price from a fictitious to a popular basis. Dr. Taylor describes the following species: Lactarius deliciosus. Canthaxellus cibarius, Marasmius oreades, Hydnum repandum, Agaricus campestris, Coprinus comatus, Morchella esculenta, Clavaria cinerea, Clavaria rugosa, Boletus edulis, Lycoperdon giganteum, and Fistulina hepatica. All these species are shown in colored lithographs and described sufficiently for their identification in the text, in which, moreover, directions are given for gathering, preserving and cooking each one. In an appendix the reader is instructed how to cultivate mushrooms, and Mr. Falconer's description of a new species, Agaricus subrufescens, is quoted.



CRYSTALS OF TARTRATE OF SODA.

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THE AMERICAN

MONTHLY

MICROSCOPICAL JOURNAL.

VOL. XV. OCTOBER, 1894. NO. 10.

On the Limitation of Tuberculosis.

BY DR. W. W. ALLEGER,

WASHINGTON, D. C.

[Read before the American Association of Microscopists, August, 1894.]

The past twelve months have marked an era in the prophylaxis or prevention of consumption. There no longer exists any reasonable doubt as to the infective nature of this disease ; and, being communicable, it follows that it is to a greater or less extent preventable. There is, therefore, no more important subject to which scientists may devote their attention and enlist their services than the consideration and adoption of measures looking to the control and limitation of tuberculosis : since it causes more deaths than any other disease, being directly responsible for one-seventh of the entire mortality of the globe and fully one-fourth of the deaths which occur between the ages of 20 and 40 years. The task, however, is not an easy one. Not that we lack knowledge or means to wage a fairly successful warfare, but because the evil is so wide-spread and deep-seated and mankind has become so inured to its ravages and has so long regarded it as hereditary and unavoidable that it is very difficult to overcome the apathy thus engendered and secure the vigorous and efficient co-operation of all concerned. Moreover, being an essentially chronic disease, the measures necessary in its control are irksome and hence hard to enforce, particularly among the indifferent and incredulous. If one-tenth part of the number of deaths occurred in any given community from some such acute

disorder as Asiatic cholera, yellow fever or smallpox as annually occur therein from tuberculosis, there would be such a popular uprising as to insure the adoption of every known measure for its control or eradication; but against the insidious inroads of this familiar scourge, with all the long standing and firmly fixed prejudices and erroneous ideas to combat that must first be overcome, the case is otherwise. As it is, it is necessary to educate the masses as well as to enlist the services of the medical profession, and to secure the adoption by national, state and municipal authorities of such regulations as are necessary to accomplish the end in view. This is what advanced writers and workers of the day are endeavoring to do.

While many are seeking diligently after a specific for the cure of tuberculosis, and while we know that it is cured in a much larger per cent of cases than was at one time supposed, others, recognizing the difficulties yet to be encountered in the management and cure of the disease, and believing that it is more blessed to prevent than to cure, have turned their attention to questions of prophylaxis. And, thanks to Robert Koch and his co-laborers, the advances that have been made in our knowledge of the nature and mode of propagation of tuberculosis since the discovery of its essential cause—the "bacillus tuberculosis"—we are in a position to institute measures which, if faithfully carried out, cannot but greatly diminish and perhaps eventually stamp out this monstrous scourge.

WHAT THE BACILLUS IS.

The first essential to success in prophylaxis is a clear understanding of the biological character of the infectious agent, the conditions under which it is capable of setting up morbid processes and the channels of infection.

The bacillus tuberculosis is a minute rodshaped veget-

able organism from 1.5 to 3.5 micromillimeters in length, and about 0.2 of a micromillimeter in breadth. It is recognized by its morphology and staining peculiarities, the latter being so distinctive as to permit its ready differentiation from all other bacteria, with but a single known exception.

The bacilli are found in the sputum of persons suffering from pulmonary and laryngeal tuberculosis; in miliiary tubercles and fresh caseous masses in the lungs or elsewhere; in recent tuberculous cavities in the lungs; and in the milk of a considerable per cent of tuberculous cows. Except in the laboratory, where a suitable pabulum,proper temperature and other surroundings are applied, the tubercle bacilli are incapable of living a saprophytic form of existence, and therefore do not multiply outside of the body of infected persons or animals, although many recent observations have shown beyond cavil that they retain their vitality and virulence for months and even years in rooms that have been inhabited by tuberculous persons, particularly in crevices and dark recesses, where they are protected from the action of strong rays of light. Exposure to the direct rays of the sun, however, destroys them, in a few minutes or hours, according to the thickness of the layer of sputum or other material with which they have been deposited, while a bright diffused light also renders them inert after a variable, but much longer time. The bacilli gain access to the organism in some instances through the milk and meat of infected animals, in which cases they are capable of setting up, in susceptible persons, especially in children, tuberculosis of the bowels; fresh wounds and abraded surfaces also occasionally become inoculated; but in the vast majority of cases their portal of entry is through the respiratory organs, by the inhalation of dust containing the bacilli or their spores.

HEREDITY DOUBTED.

The popular idea that tuberculosis is generally inherited cannot, from a bacteriological standpoint, be subscribed to. While we do not say that the direct transmission of the disease from parent to offspring is impossible, yet we do affirm that this is at least of comparatively infrequent occurence. It is true that a specially vulnerable or invulnerable type of tissue may be and is inherited, the former rendering the individual prone to infection, the latter conferring partial or complete immunity from infection by the bacillus tuberculosis. In the one case the seed, when sown, falling on good ground, bears fruit not an hundred, but many million fold ; in the other, falling by the roadside or on stony ground, either fails of growth altogether or develops to a limited extent only.

HOW THE BACILLUS GROWS.

Having once gained access to vulnerable tissues whether that vulnerability has been inherited or acquired, the irritation set up by the growth and multiplication of the bacilli, aided by the poisonous chemical products elaborated in their cells and from the albuminoids in the tissues of the host, little nodules or new growths are developed which constitute the essential lesions of tuberculosis. These new growths or tubercles, as they are called, have a tendency to undergo caseous degeneration and break down, and the detritus, containing large numbers of the bacilli, is thrown off with the sputum in cases of pulmonary tuberculosis. If the sputum thus contaminated is allowed to soil the floor and become dry and pulverized, the air of the room, laden with dust bearing the spores of the bacilli, becomes a source of danger to the non-tuberculous persons in the apartment, as well as the patient himself, whose chances of recovery are diminished by reason of constant reinfection through germs which nature has once expelled.

The bearing of these facts upon the question of prophylaxis seems almost too obvious to need special mention, this portion of the subject might be here dismissed but for the fact that many still entertain the idea that the relation of the tubercle bacillus to tuberculosis is a matter about which there exists much doubt, the claims in the affirmative being believed by them to rest chiefly upon the opinions of over-zealous or impractical bacteriologists; and for the reason that certain authorities have recently come forward with the statement that tubercle bacilli bear no more relation to tuberculosis than do certain other micro-organisms; that we may and do have tuberculosis, without the presence or intervention of the tubercle bacilli; and that in carrying out measures of prophylaxis against tuberculosis disinfection may be safely disregarded.

The truth is that no question in general pathology rests on a firmer foundation than does the fact that the tubercle bacillus is the essential cause of tuberculosis. Not only are those bacilli constantly present in this disease, and absent from non-tubercular lesions, but when isolated in pure culture, even through several generations, and then introduced into the bodies of susceptible animals, they are capable of causing tuberculosis in the creature thus inoculated. This is considered to be a crucial test, and has been successfully performed over and over again by scores of competent observers in several different countries.

NO CONSUMPTION WITHOUT INFECTION.

There are many causes which may predispose an individual to the development of tuberculosis; but no one of these, nor all of them combined, will suffice to determine an attack of this disease unless the bacillus tuberculosis is super-added. Lesions simulating tuberculosis may be induced, but not the true disease. A minute

quantity of material from one of these pseudotubercles introduced into the body of a susceptible animal will not give rise to tuberculosis; while if an equal amount of material from a true tubercular nodule be introduced, the result will be tuberculosis in the inoculated animal.

In addition to this experimental data, clinical evidence has been accumulated which is deemed sufficient to establish the communicability of tuberculosis. And, if communicable, how shall we explain the power of tuberculosis to propagate itself, in the light of our present knowledge, except upon the theory that the disease is due to a *contagium vivum.*

In response to a question as to contagion, asked by a committee of the British Medical Association, 261 replies in the affirmative were received, among which were 158 cases of supposed contagion through marriage. Of these, Weber's cases are of special interest. One of his patients lost four wives in succession; one three; and four, two each.

PROOF OF ITS COMMUNICABILITY.

Another clinical observation of interest is the fact that patients received in the wards of general hospitals with tubercular patients sometimes contract tuberculosis during their stay and are discharged cured of the original malady only to find themselves victims of tuberculosis; or, if their stay has been more protracted, remain only to succumb to the latter disease. Biggs calls the attention to the frequent occurrence of such cases in the hospitals of New York, and the writer has observed a few similar instances in the hospitals of Washington.

Mortality statistics show the influence exerted by occupations requiring confinement in dust laden and crowded workshops. In every thousand deaths from all causes in farmers only 103 are due to pulmonary tuberculosis; in fishermen, 108; in gardeners, 121; in agricultural labor-

ers, 122; and in grocers, 167; while in tailors the number reaches 290; in drapers, 301; in printers and compositors, 461, or nearly 50 per cent. Among the Cornish miners the number is said to exceed 60 per cent. In prisons and other public institutions where insufficient ventilation and exercise, lack of variety in food, and overcrowding are combined with infection of the apartments the death rate from tuberculosis ranges between 40 and 50 per cent.

Cornet's observations are interesting and instructive. Of 118 samples of dust collected from the walls and bedsteads of hospital wards and the rooms of phthisical patients, 40 were infective and produced tuberculosis on being inoculated into susceptible animals. With the 29 samples collected from localities only occasionally visited by consumptives the results of inoculation were all negative. Virulent bacilli were obtained in 15 out of 21 medical wards. In two wards containing several consumptives the results were negative.

BEDBUGS AS SOURCES OF INFECTION.

Among the novel sources of infection are bedbugs. Dewevre relates an example. Tuberculosis having occurred in a young man occupying the bed of his brother, who had previously died of this disease, but in a room thoroughly disinfected, the inefficiency of the disinfection was explained when an inspection of the corpse showed that it was covered with the bites of bedbugs, and that his bed, which had escaped disinfection was filled with these animals. Thirty of the bugs were gathered and inoculated in three guinea-pigs, which soon died of tuberculosis. Sixty per cent of the bugs were found to be tuberculous. In the other series of experiments the bedbugs were placed in contact with sputum and some weeks afterwards virulent cultures were obtained from them. If these bugs can transmit the disease from one

to another, they may furnish the missing links in cases not otherwise explicable.

In view of all the facts its seems idle to deny an etiological role to the bacillus tuberculosis, and pernicious to omit measures looking to its exclusion and destruction in connection with the prevention of tuberculosis and the care of those suffering from this disease, notwithstanding the statements of a few recent writers as to the needlessness and inutility of such measures.

DISINFECTION OF FIRST IMPORTANCE.

In our opinion the disinfection of the sputa and other infectious materials, and the destruction or proper disposal of tuberculous cattle are of the very first importance. If this could be thoroughly done there is no question but that this monstrous scourge could be eliminated from the face of the earth. But while the hope of complete eradication cannot now be entertained, we may at least hope for as much success as has been attained in limiting the spread of smallpox, cholera and other infectious diseases. We do not underestimate the importance of measures of general hygiene, and are not unmindful of the fact that while tuberculosis is communicable, it is so to a much less extent than measles, scarlet fever, smallpox, etc., to which the term contagious is more properly applied, and we are aware that it generally requires for its development long and close contact with the sources of infection, or, in addition to exposure, the operation of some depressing agent or source of irritation sufficient to overcome the natural resistance of the organism and render the tissues vulnerable. For this reason special care should be exercised to avoid all possible sources of infection during the existence of disorders of the respiratory organs or convalescence from any of the exanthemata, whooping cough, diabetes, or other diseases likely to lay the foundation for the development of tuberculosis.

HOW IT SHOULD BE DONE.

Persons suffering from tuberculosis should not be permitted to work in close apartments with other workmen, both because their presence in such places constitutes a grave source of danger to others and for the reason that their own chances of recovery under such conditions are very materially lessened. Nor should they continue the pursuit of other occupations in which they are likely to infect others.

Separate hospitals should be erected for the treatment of tuberculosis as it occurs among the poor, and these should be erected in dry, healthy locations without the city limits, surrounded by spacious grounds, and the rooms well ventilated and an abundance of sunshine admitted.

Rooms vacated by tuberculous patients should be disinfected before being reoccupied.

Linen worn by tuberculous patients should be placed in boiling water when changed, and other clothing, if accidentally or otherwise soiled with sputa or other infectious material, should be disinfected by exposure to steam, hot air or formalin vapor or solution.

Table-ware used by tubercular patients should be placed in boiling water for a few minutes before being washed with other dishes.

Kissing should be avoided.

Tuberculous mothers should not nurse their infants.

Dairies and abattoirs should be rigidly inspected by competent agents appointed by the state, municipal or national authorities and all diseased milch cows and infected meat should be promptly condemned and properly disposed of. Meat should be thoroughly cooked and never be eaten rare, unless the animals slaughtered are known to have been free from tuberculosis. Milk coming from dairies that have not been properly inspected should not be used, especially by young children, without first

being boiled or, what is preferable, the quality of the milk being considered, kept at a temperature of 70 C. for half an hour.

PRECAUTIONS FOR PATIENTS OUT OF DOORS.

When on the street or away from home the sputa should be received in paper napkins and deposited in a tight metal box carried for the purpose. Immediately on arriving home the napkins should be burned and the box disinfected by heat, an efficient disinfectant solution or even by boiling water. Handkerchiefs should not be used to receive sputa.

Isolation or quarantine is not necessary. The expired air from the lungs of the sufferer from tuberculosis *does not contain the germs of the disease,* and there is no danger to the most intimate associates so long as contact with the sputum is avoided.

WHAT IS BEING DONE.

It is pleasing to note that sanitary authorities throughout the world have already taken measures looking to the restriction of tuberculosis. The Government of Prussia has published a series of recommendations for the prevention of this disease. The Minister of the Interior at Wurtemberg, and the Minister of the Interior of Russia have issued instructions regarding measures to be taken against the spread of tuberculosis in public institutions. The Michigan State Board of Health has officially declared it a contagious disease, and included it among those in which compulsory notification is required.

The New York City Health Department has determined to inaugurate advanced measures for its active surveillance, and the Department of Charities and Correction of New York has at the same time signified its intention of setting apart a hospital for the exclusive treatment of consumption as it occurs among the poor.

The Board of Health of Philadelphia has issued a cir-

cular showing the number and percentage of deaths from tuberculosis as contrasted with other infectious diseases, stating the chief sources of infection and giving full directions as to the disinfection of the sputa, table-ware, clothing, and the rooms occupied by tubercular patients. And the city has established a large plant for the proper disinfection of clothing. In this at least, one of the public hospitals (Freedman's) has caused the segregation of the cases of tuberculosis and adopted other proper measures, including disinfection of the sputa and wards, to prevent the inoculation or infection of other patients.

The State of New York has enacted a law requiring the inspection of cattle and the destruction of those found to be diseased. One-half the value of the condemned animals is paid to the owner by the State, and, in case the autopsy reveals no tubercular lesions, the full value.

On "Species" in the Desmidiæ.

By ARTHUR M. EDWARDS, M. D.
NEWARK, N. J.

What are known as "species," that is say forms which are thought to be distinct one from the other I do not believe exist. We must distinguish forms by some other mode, by physiology for instance. By that method they grow, and this I have attempted to make plain in the Bacillariaceæ, a group of the Protista. Another group of the Protista now claims our attention. With the Desmidiæ of course I am not so familiar as with Bacillariaceæ.

I have had growing in a large jar, this spring, some aquatic matter which I gathered when the frosts of late winter disappeared, and I have been studying it long and often. In it I found Ankistrodesmus falcatus J. R., a slender Desmid, green and looking like a straight shuttle, that is to say sharply pointed at each end, sometimes straight, sometimes sigmoid and 1-550 in. long and

1-7353 in. broad. It has been called Raphidium fasciculatum by F. T. Kutzing, in his "Species algarum," 1849. He gives the synonyms of this form thus: Binatella calcitrapa, A. de B. Alg. Fal. Pl. VIII.; Closterium gregarium Menegh.; Xanthidium (?) difforme C. G. E. Infus. p. 147, Tab. X, Fig. XXVI.; Staurastrum falcatum et paradoxum Ehrenb.; Ankistrodesmus falcatus A. de B. in list, 1845; Micrasterius falcata Corda, and Closterium falcatum Menegh.

It is like a Closterium, being elongated and attenuated but aggregated into families forming fascicule or faggot-like bundles. This is the way they appear commonly, but mine are single. Then there is a form, called Ankistrodesmus convolutus Corda, in which the cells are much curved, cresent shaped, somewhat rapidly attenuated with the ends subacute. But A. falcatus passes into that by imperceptable degrees. There is another form called A. contortus in which the cells are arcuate or sigmoid, somewhat gently inflated at the centre and the ends are drawn out long and very fine. Now these three forms, or species as they are called, can hardly be distinguished. Which can we say are arcuate or sigmoid, slender or not attenuated ? In my form, I see the passage from one into the other and they are not aggregated into faggot-like bundles but separate and swimming about in the water, very slowly it is true. So I shall make it into the form, for I am indisposed to call them species of falcatus. And as to the genus Ankistrodesmus, I cannot see how the growing in faggot-like bundles is distinctive for they do not so grow. They must be placed in Closterium P. A. C. Nitzsch. But the genus of these forms I do not now wish to go into. I merely wish to place the form in the genus Closterium falcatus A. de B. and described as above. I call attention to these beautiful atomies, the Desmids, so as to show that they will warrant study.

A Study of the Microscopic Phenomena of Commencing Inflammation, With Special Reference to the Diapedesis of the White Blood Corpuscles.

BY CHAS. F. CRAIG, M. D.

DANBURY, CONN.

Away back in the olden times, observers noticed that a majority of the diseases of man were accompanied by certain symptoms and these were constant and formed an unbroken chain in many instances. They were: swelling, redness, pain and heat in the diseased part or organ, and also, at times, an increase in the pulse-rate and a rise in the general temperature of the body.

We have no difficulty in recognizing here the characteristics belonging to inflammation and fever. The tissue which was red, swollen and painful was said to be inflamed, while if there were an increased pulse-rate, higher temperature, and a change in the chemical activities going on within the body, there was said to be fever.

The connection between inflammation and fever was generally easily seen, but sometimes one occurred without the visible presence of the other and their connection was obscured, but there came a day when the microscope was introduced into medical research, and then the production of inflammation and fever were closely studied and light was shed on what was before darkness.

By careful experiment the following facts have been clearly proved regarding the origin of inflammation.

First, there is, on irritating the tissue experimented upon a dilatation of the capillaries, then a migration through the capillary walls of leucocytes or white blood corpuscles, which spread into and increase in the surrounding tissue; then a blood stasis in the dilated capillaries and a congestion in the inflammatory area. These are accompanied by disturbances in the nutritive processes and a heightened temperature, *i. e.* fever.

Such, then, are the changes which characterize the commencement of inflammation and its progress as seen under the microscope.

With the thought in my mind that "we learn new things only by more carefully and patiently observing old ones," I have spent nearly all of three months (July, August and September) in studying, beneath the microscope, the following phenomena of commencing inflammation—dilatation of the capillaries, slowing of the blood stream, and the escape of the leucocytes or white blood corpuscles from the blood vessels into the surrounding tissues.

I made these observations with but little hope of discovering any new facts where such observers as Colmheim, Lister, Stricker, Bottcher and others had so carefully and scientifically worked, but simply to convince my own mind of the truth of their revelations, and to decide, if possible, to my own satisfaction, certain points, regarding the diapedesis of the leucocytes, which are still in dispute. It will hardly be necessary for me to say that my results confirmed their observations, and in one or two points, my observations, although differing somewhat from theirs, have cleared up, to my own mind, disputed questions, the principal of which is that regarding the part taken by the leucocyte in its passage through the vessel wall.

I put forward no new theories, but simply state, in as plain a manner as possible that which I have seen only, leaving to others the task of working out the theory which will accord with the facts.

My observations have been made upon the foot-webs and mesenteries of ten frogs, and extended over a period of about three months, as before stated. I have not attempted to write out a description of each individual case for that would be but repetition, but simply have drawn general deductions from them all.

METHOD.

The method I have followed in making my observations is simple in the extreme. First, I constructed a platform for the stage of my microscope, on which to place the frog, consisting of the parts to be mentioned.

I took a piece of cork, twelve inches long, four inches in width, and half an inch in thickness at the center of its long diameter and an inch from its side I cut a hole two inches long and about an inch wide; over this I placed a glass slide, such as is used in microscopy, sinking its two ends even with the surface of the cork. The slide entirely covers the hole cut in the cork.

A quarter of an inch from the outer edge of the slide, I cut a groove in the cork running a little beyond the ends of the slide and parallel with it, and a quarter inch from each end of this groove, another groove at right angles to it, which is connected with the rubber pipe to allow for drainage, if needed. The groove is intended to contain the intestine.

This apparatus is simply fastened upon the microscope stage, seeing that the glass slide covers the aperture in the stage, and the frog arranged upon it, after being curarized by injecting a little of a watery solution of the poison in the dorsal lymph sac.

If the web of the foot is to be examined it is simply stretched across the slide and held in place by felted pins, placed between the toes but not piercing them.

If the mesentery is to be examined, the frog is placed on the apparatus after being curarized, the abdomen carefully opened and the intestine gently lifted out with felted forceps and placed in the groove, thus stretching the mesentery smoothly over the slide where it can be easily examined. My method was to get a fresh frog from a pond near by each time I needed one.

On the first ten frogs I made no observations whatever of the mesenteries but only of the foot webs, while on the

last ten I observed the mesenteries only. I tried to observe as carefully and as long at a time as I could, and I would say here that it is only by watching this process of inflammation from the time it commences until we can no longer follow it with our microscope, without once taking our eyes from it, can we hope to see and understand all of its phenomena; but in my own case I found it intensely tiresome to observe longer than eight consecutive hours at a time so that I probably missed much that I might otherwise have seen had I had greater endurance and patience.

EXAMINATION OF FOOT-WEBS.

I examined the foot web of a frog with a medium power say a $\frac{2}{3}$ in. objective. The first thing noticed is the great number of vessels and capillaries, filled with a swiftly moving current of blood; so swiftly, indeed does it move, that the constituents of the blood cannot be distinguished, and we see it only as a never ending motion, without known composition.

At various situations in the field beautifully branched pigment cells are seen and on carefully focussing we can distinguish the cells of the surface epithelium, each cell containing a barely distinguishable nucleus. This is the appearance of the web before it is acted upon by an irritant, and such will be the appearance for hours, if the part is kept moistened and not irritated.

But on touching the web lightly with nitrate of silver, or even scratching it with a needle, we get an entirely different appearance, and the process of inflammation begins. At once there is a dilatation of the arteries, this dilatation extending gradually to the veins and capillaries. As far as I could determine this dilatation was not preceded by any contraction, and affected the arteries mostly, and the capillaries but slightly, although the ratio may have been alike. This dilatation seemed to in-

crease steadily for about six hours, although Cohnheim states that it steadily and slowly increases for twelve hours. At the beginning of this dilatation there was an acceleration in the flow of the blood plainly noticeable, but this, after lasting only about an hour, resulted in a considerable retardation of the flow, the vessels still remaining dilated.

After an hour and sometimes in the smallest capillaries, much sooner, pulsation became evident and the current of blood so slow that in the small veins and capillaries, the individual corpuscles became distinguishable. They can first be distinguished in the veins. In two of my observations, this dilatation took place and subsided without any of the other phenomena of inflammation occuring, but, afterwards, a second dilatation with slowed stream came on slowly, which was constant, lasting as long as the cause operated. This last dilatation was, no doubt, the vascular change of the inflammation due to the irritation.

Returning to the time when the corpuscles are first to be distinguished, the following phenomena were observed, useing now a one-sixth objective. As the blood stream became slower, white corpuscles or leucocytes were seen in the plasmatic layer in the smaller veins, rolling along sticking here and there, coming to a stand-still for a few seconds, then rolling on again, and at last often sticking fast to the vessel wall, resisting all attempts of the current to dislodge them.

A vessel at this time presents the following appearance, the white corpuscles being scattered here and there along the vessel wall, while the central canal of the vessel, enclosed on each side by the plasmatic layer in which the leucocytes lie is filled with the red corpuscles which can be distinguished very readily as the stream gets slower and slower. One of two things now generally occurred, either the leucocytes continued to accumulate on the

vessel wall, or else the vessel became so thickly packed with corpuscles as to present the appearance of a red injection mass, all movement ceasing. The contents of the vessel sways forward and backward with the impulse transmitted to the stream by the beating of the heart. This is known as the stage of oscillation and is succeeded by a complete stagnation, called stasis, in which no movement of any kind occurs.

I have spoken of the accumulation of leucocytes along the vessel wall, as though in this last instance it did not occur; by this, I mean that in my observations the vessel filled so rapidly that the accumulation of leucocytes was hidden from view, although if the field thus obscured be watched long enough, the leucocytes are found congregating outside the vessel, thus showing that diapedesis still goes on.

When, however, the blood stream still continues to move on, the leucocytes accumulate more and more, until the vessel wall in some cases, becomes lined with them, often two or three cells in thickness.

If such a small vein be watched very carefully for a varying space of time, from half an hour to several hours, the following phenomena are seen to take place. (It is not necessary, however, that the vessel wall be lined with leucocytes in order that these phenomena take place, for in all those observations in which I obtained the clearest and best view of the process, the leucocytes were only situated here and there upon the vessel wall.)

As the vessel is watched, the leucocytes are seen to gradually penetrate its wall, and finally to emerge upon the other side, having passed entirely through the vessel wall into the surrounding tissues.

This process seems to be divided into four distinct acts. At first the leucocyte is seen to adhere to the vessel wall; then it sends a process into and through the wall; following this, the remainder of the cell is drawn after or fol-

lows this process, at one time in its passage appearing constricted in the middle, and finally, the leucocyte passes completely through the wall, and moves into the surrounding tissue, by virtue of its amœboid movements.

In these four acts, the process, known as the diapedesis of the white blood corpuscles or leucocytes consists, and it is said to be due partly to the independent motility of the leucocytes themselves and partly a filtration of the colloid mass of the cell by the force of the blood pressure, and probably taking place through the spaces between the endothelial cells, the vessel wall itself, having been altered in some way by the process of inflammation.

In my observations, I very carefully watched all of the phenomena and could observe nothing peculiar during the first stage, *i. e.* adherence to vessel wall, nor during the passage of the leucocyte through the wall, but in the second stage, *i. e.* when the cell sends out its process into the wall, I observed a strange thing, namely, that the process sent into the wall by one cell was almost identical with the process that any other cell sent out, both in size and general outline.

This process was a cone shaped, rather pointed one, which seemed to work its way into the wall of the vessel and, in fact, it seemed to quietly and steadily push its way on and in, until at last it emerged upon the outer side of the vessel. I also noticed in every case, which I observed, where, after adhering, the leucocyte was swept from its hold and carried on in the current, that it retained its penetrating process, and did not, as long as I could see it regain its irregularly circular shape.

From these two facts, it seems more than probable that this penetrating process is due to the activity of the cell itself, rather than that it is such a one as would be produced by the blood pressure acting upon the soft colloid material of the cell and pressing it into the vascular wall, for a process so formed would disappear as soon as the

leucocyte was swept off into the current. Again, the uniformity of the shape of the process tends to favor the view that the leucocyte penetrates the vessel wall mostly by its own independent power of locomotion.

Now, as the leucocytes penetrate the vessel in greater and greater numbers, as serum accumulates also, the surrounding tissues become infiltrated and the field observed through the microscope becomes covered by a false membrane, composed of leucocytes connective tissue cells, fibrine and serum.

The above facts constitute the phenomena of inflammation which I observed in the web of the frog's foot. I did not carry my observations further, as I mostly wanted to study the diapedesis of leucocytes, and I found that owing to the amount of material in the field, other than blood vessels, *i. e.* pigment cells, epithelial cells, etc., I could not obtain a good clear view of the process, and I accordingly conducted all my other observations upon the mesenteries of frogs, and obtained, I believe, a much more accurate view of every step of the process, as far as a microscope will reveal it.

From the foot-web of the frog then, I observed, besides the process usually described, *i. e.* the adherence to and penetration through the vessel wall, of the leucocyte, two distinct and characteristic phenomena.

1. The process, which any leucocyte sends into the vessel wall in penetrating it, is almost identical, in size and shape, with that which any other leucocyte sends off in the same act, or in other words; the leucocytes all send off similar processes into the wall of the vessel before penetrating it.

2. In all the cases which I observed, if a leucocyte, after sending off such a process, is swept from the vessel wall by the current, it always retains that process and does not return to its former shape, providing it had really began penetrating the wall.

OBSERVATIONS ON THE MESENTERY.

At this point, I transferred my observations to the mesenteries of frogs, instead of the foot-webs, but before doing so I made a thorough examination of the corpuscles of the frog's blood with the following results.

Besides the oval red corpuscles I found that there are two distinct kinds of white corpuscles present in the blood which may be described as follows:

The most numerous variety are large, about 1-1200 in. in diameter, finely granular, irregular in outline, having fine projections from its surface and having a two, but generally three part nucleus, surrounded by finely granular protoplasm.

The second variety is less numerous, more coarsely granular, smaller, and contains one nucleus. The granules in the protoplasm are sometimes seen to rush from one side of the corpuscle to the other. The corpuscle exhibits active movements which are, of course, amœboid, as does also the larger variety. Unfortunately, when watching the process of inflammation, I could not distinguish the difference between these two varieties for the following reasons.

1. In a small vessel white corpuscles are not present in sufficient numbers to detect the two varieties, and when present in large numbers, so crowded together as to make it impossible.

2. The nuclei of the white blood corpuscles cannot be clearly made out without the use of reagents, which, if used, would interfere with the process of inflammation.

I found it necessary as stated in order to clearly define the nuclei and to see their number to add to the blood a little dilute acetic acid, which clears up the surrounding protoplasm, and brings the nuclei very clearly into view. If desirable, magenta may now be added which will stain the nuclei very beautifully. Adding dilute alcohol will also bring the nuclei into view.

Having now thoroughly examined the frog's blood, I turned my attention to the mesentery which was to be my field of operation. This is seen, by the microscope, to be composed of the following elements, being a most typical example of all serous membranes.

It is transparent, and composed of—

1. Very fine delicate connective tissue fibrils.

2. Connective tissue cells.

3. Some elastic fibres, and it is traversed by a beautiful net-work of lymphatic and vascular vessels, and channels. In order to watch for a long time, the process of inflammation in the mesentery, it is necessary to keep it moistened by a weak salt solution ($\frac{3}{4}$ per cent sol.). Carefully opening the abdomen of the frog and spreading the mesentery upon the microscope stage, as described before, I observed the following phenomena.

A slight inflammation is caused by simple exposure of the mesentery to the air, and there is a slight dilatation of the arteries with an accelerated current. At this stage we get an appearance as follows :

The vessels show beautifully lying in the transparent membrane, with no pigment or other matter to obstruct the view.

The stream of blood is so rapid that it is impossible to distinguish the individual corpuscles, but gradually, the irritation being kept up, in from four to five hours, sometimes much sooner, if a medium sized vein be watched, the stream begins to grow slower, and soon the corpuscles become distinguishable, first in the smaller vessels.

This slowing occurred sooner in the mesentery than in the web of the foot, and more rapidly the greater the irritation. As the stream becomes slower the same phenomena are observed as in the web, namely : the red corpuscles occupy a central position in the vessel, there being on each side of this stream of red corpuscles, a zone of plasma, in which most, but not all, of the white cor-

puscles are moving. This is said to be because of their less specific gravity, for in 1868, Schklarewsley showed by physical experiments, that particles of least specific gravity, in glass capillaries, are pressed toward the wall, while the ones of greater specific gravity, remain in the center of the stream.

In the smallest capillaries the red corpuscles move along in single file, sometimes being compressed between the walls but regaining their former shape, on account of their elasticity, on reaching larger vessels. The white corpuscles are plainly seen, rolling along, sticking here and there on the vessel wall and sometimes being swept off again by the axial current.

First. After a leucocyte has remained for a variable length of time upon the vessel wall, and just before it can be seen to send a process into it, the granules, which I stated as being distributed throughout it, all seem to be in motion and congregate thickly at that portion of the leucocyte lying furthest from the vessel wall or in other words, nearest the axial stream of the vessel.

Second. The leucocyte next sends off a process, which is almost exactly the same as that which every other leucocyte sends off, in its general outline, and this process is entirely free from pure protoplasm. This process seems to gradually push or work its way through the wall.

Third. Now occurs a most singular thing. As the vessel wall at last gives way and the process of the leucocyte emerges upon the outer side of the vessel, the granules, which have, during the penetration of the wall, congregated at the furthest part of the leucocyte from the wall,—these granules fly swiftly through the passage thus made in the vessel wall by the protoplasm of the cell, from the interior of the vessel to the furthest point of the process outside the wall, leaving the part of the leucocyte not yet through the wall, free from granules.

This now passes through and the leucocyte regains its former shape, the granules becoming distributed over its surface.

In several instances, while the leucocyte was in the act of penetrating the vessel wall, it was swept from it by the blood stream, but the granules still held their position in the part of the cell away from the penetrating process, and more than this, in one case a leucocyte so swept away, adhered again to the wall, while still in the field, and penetrated it with the self-same process, passing through all the stages mentioned.

These phenomena which I have just mentioned indicate to my mind that the leucocyte penetrates the vessel wall largely by its own cell activity, and I am convinced that such is the fact.

As the blood stream becomes slower and slower, the process is obscured by the accumulation of leucocytes, and when at last the current stops, the vessels become filled with them, and diapedesis can no longer be watched. To see the process the best, a point in the vessel where the blood stream is quite rapid and the leucocytes few in number, is most favorable.

To sum up then, the following facts are those which I think are most important and which I have not as yet seen any other account of.

1. The leucocytes, in penetrating the vessel wall, send off processes similar in shape and size.

2. If a leucocyte, while in the act of penetrating the vessel, was swept off into the blood current it retained its process as long as I could see it.

3. The granular matter in the leucocytes becomes arranged in the portion of the cell furthest from the penetrating powers so that the process contains only clear protoplasm.

4. If the leucocyte be swept away while penetrating, the granules still retain their position, and in one case

the leucocyte was seen again to attach itself to the wall, and use the same process in penetrating it.

5. The clear protoplasmic process works its way through the wall, and in the channel of protoplasm thus formed, the granular matter rushes through the wall and occupies the clear process which is now outside the vessel, and leaves clear the portion of the leucocyte still within the vessel.

6. This clear portion soon comes through the wall and the leucocyte regains its former shape.

The thought came to me that perhaps it was the coarsely granular corpuscles, heretofore described, in which the granules are seen to rush from one side of the cell to the other, that were the ones which penetrated the wall, but when I considered the small number of this variety when compared with the finely granular leucocytes, and the large number of leucocytes which so escape, I thought that it probably must be the finely granular which constituted the larger part of the escaped leucocytes.

Of course, the phenomena observed give rise to many interesting questions, but to me it seems evident that the condition brought about by irritation causes in the leucocyte, a certain activity which is manifested by the phenomena, and which results in its escape from or rather through the vessel wall. Whether it takes place through the so-called stomata, or through the cement substance between the cells (endothelial) of the vessel wall, I could not determine.

In conclusion I would say, that although my results differ somewhat in some particulars, from those heretofore obtained, still, in the words of Claude Bernard: "In physiological studies we must always carefully note any fact that does not accord with received ideas, for it is always from the examination and discussion of this fact that a discovery will be made if there is one to be made."

Read at the Brooklyn meeting of the Amer. Micr. Society.

Microscopical Technique Applied to Histology.—VII.

[FROM THE FRENCH OF RENE BONEVAL.]

(*Continued from page 270.*)

Carmine mass.—This furnishes beautiful injections, but the difficulty in exactly neutralizing it without precipitating the carmine makes the Prussian blue preferable for every day work. . . .

Liquid masses.—In certain circumstances it is advantageous to use a cold liquid mass. The saturated solution of Prussian blue acts well; a solution of silver nitrate (1 to 300) may be used to impregnate the endothelium of the vessels.

INJECTING.

We advise beginning with the frog.

Injecting with the blue gelatine.—Having immobilised a large frog by piercing the medulla oblongata, cut the sternum on a level with the xiphoid appendage, making the incision parallel with the long axis of the body; open the pericardium, lift the heart out of the thorax and cut off the point with scissors. Put the frog in warm water (40°C) and wait for the blood to run out. Fill the syringe (which has been warmed in a water-bath), with the injection mass, put on a small canula and force out the air by pushing in the piston while the syringe is held upright. The frog being now almost bloodless, introduce the canula into the heart up to the aortic bulb and place a ligature around the latter to hold the canula in position. Make the injection gently. When the vessels are full (it will then be difficult to push the piston), place a ligature beyond the canula. . . . Put the frog in a large vessel of two per cent of ammonia bichromate solution. In a few hours, the gelatine being hard, open the abdomen to allow the liquid to penetrate more rapidly. Cut sections after hardening in alcohol and imbedding in gum. The preparation should be mounted in dammar.

[It may be well to repeat that M. Boneval always refers to dammar when he mentions a resinous medium. This has hitherto been translated by the word balsam. The reader may use either dammar or Canada balsam].

Injecting a frog with silver nitrate.—Prepare the frog as described, omitting the warm bath ; . . . .use a solution of silver nitrate, 1 to 500. . . . Open the frog in a basin full of water and expose to direct sun light. We advise the examination of the mesentery, the lungs and the bladder.

The mesentery should be spread on a slide, partly dried and mounted in balsam (*dans la resine*) ; the lung and the bladder should be opened in distilled water, brushed to remove the epithelium, and mounted in balsam, the inner face upward.

Injecting a mammal with the blue gelatine.—Select the rabbit or the rat. The former may be injected by the carotid or by the crural artery, after being killed by piercing the medulla oblongata. . . . Cut away the hair with scissors, sieze the skin between thumb and fingers on a level with the upper end of the trachea. The horizontal fold thus formed is incised vertically. Raise the aponeurosis between the median line and the border of the sterno-cleido-mastoid and slit it with the scissors. Enlarge the cut with the grooved probe, dissect away the muscle so as to show the carotid, the vein and the three accompanying nerves. . . . Open the sheath with a blunt hook and isolate the carotid. To expose the crural artery of the rabbit is easy. Cut the skin in a straight line from the middle of Ponpart's ligament to the inner side of the knee ; the naked sheath of the vessels is seen and the artery somewhat behind and between the vein and the nerve ; remove the cellular tissue and expose a portion of the artery. . . . Insert the canula through a V-shaped cut, tie it in place, inject, put the rabbit in cold

water for 5 or 6 hours to harden the gelatine, remove the organs to the bichromate solution.

It is not impossible, but difficult, to inject a rat through the carotid or the femoral arteries.... With strong scissors cut the rat completely in two transversely through the middle of the thorax. When the blood has ceased to flow, put the lower half of the animal in a basin of warm water and introduce the canula into the aorta until its point has passed into the sub-diaphragmatic portion. Tie in place, and inject. When the mass returns through the inferior vena cava stop it by forceps, and continue the injection to the point of resistance. This method produces a fine injection of all the sub-diaphragmatic parts of the body....

LYMPHATIC GLANDS.

It is necessary to make a certain number of preparations to demonstrate the structure of the lymphatic glands.

1.—Remove a gland from man or from a dog, and put it in the $\frac{1}{3}$ alcohol. It is best to select a small gland, or to divide it into several pieces if one has at his disposal only a large specimen. In 24 hours wash in water for 20 minutes, treat with alcohol and with gum [as so often described], and section. Put the sections in water for $\frac{1}{2}$ an hour to remove the gum. Select a section, not too thin, and let it spread itself out at the bottom of a vessel of filtered water. With a marten's hair brush strike the entire surface without rubbing it.... Then turn it over and repeat the operation on the other surface. The purpose is to expell the lymphatic cells that infitrate the gland, also the endothelial cells from the meshes of the reticulated tissue. A method by which to obtain a more rapid result is to put the section in a bottle half full of water and to shake it violently. The lymphatic cells are thus freed from the reticulum without the use of the

brush. Place on a slide, stain with picro-carmine, mount in glycerine.

2.—The foregoing preparation will show the reticulated tissue freed from the flat cells that carpet it. The following method will show the same tissue with the endothelial cells. Insert the needle of a hypodermic syringe into a lymphatic gland of a dog, and gently inject a one per cent solution of osmic acid. Put the gland in water for 1 hour, follow by gum and by alcohol. Make very thin sections and shake them for some time in a bottle half filled with water.

3.—Section parallel with the long axis of a gland through the umbilicus (hilum). Harden by alcohol, imbed in gum, examine in glycerine, after staining by picro-carmine. To show the differences between the sinuses and the follicles, inject into a gland some Prussian blue in aqueous solution. Fix by ammonia bichromate and harden in gum and alcohol. Stain with hæmatoxylin and mount in balsam.

4.—The ganglionic blood vessels may be studied in the rabbit or in the rat by injecting with the blue gelatine.

LYMPHATIC VESSELS.

The large lymphatic vessels should be studied in sections made by the method recommended for the arteries. Those of small diameter can be observed with all their details, (valves, muscular fibres, etc.), in the mesentery of an animal without much fat. Spread on a slide a shred of the mesentery upon which a one per cent solution of osmic acid should act for ½ an hour. Wash, stain by picro-carmine for 3 or 4 hours, wash again; examine in balsam.

(To be continued).

Dr. Hauser.—A chair of Bacteriology is to be established at Erlangen and Dr. Hauser will probably occupy it.

The Character and Uses of Glycozone.

By Cyrus Edson. M. D.

New York City.

Glycozone is defined by its discoverer, Mr. Ch. Marchand, to be a stable compound, resulting from the chemical reaction that takes place when c. p. glycerine is submitted, under certain conditions, to the action of fifteen times its own volume of ozone, under normal atmospheric pressure at a temperature of 0°C.

The necessity of using c. p. glycerine is imperative, as a presence of the water or other foreign matter in the glycerine causes the production in the resulting compound of formic acid, glyceric acid, and other secondary products, that have a harmful effect upon animal tissues.

Glycozone has a pleasant, sweetish taste. Being hydroscopic it must be kept in tightly corked bottles, and, as long as it is kept in this condition, it does not deteriorate at a temperature of even 110 degrees F.

Antagonists and Incompatibles.—Glycozone, like peroxide of hydrogen is a powerful oxidizing agent, although its action is not as rapid or as energetic in this respect as the latter compound. Consequently, we cannot safely prescribe it combined with any other drugs or chemical substances. Contact with metalic utensils decompose it. We must therefore use glass or hard rubber vessels and syringes when administering it.

Physiological Action.—When taken into the mouth and stomach glycozone causes a feeling of warmth. It excites a flow of saliva and stimulates the gastric secretions. Being hydroscopic it attracts to itself water from the surrounding tissues but not with sufficient power to effect harm. This property is due solely to the glycerine base which enters into the composition. In very large doses, one or two ounces, it causes a feeling of distress in the epigastrium and is followed by loose, copious, watery stools, which are accompanied by severe cramps.

No effect is noted on the kidneys, the liver or the heart. Glycozone is undoubtedly slowly decomposed in the stomach, ozone being liberated and the glycerine uniting with the water from the tissues. The morbid elements with which it comes in contact probably hasten this decomposition, and in so doing

are themselves oxidized and destroyed. The free ozone in the stomach resulting from the decomposition of glycozone aids the digestive process by its presence.

Therapy.—Glycozone is, in the opinion of the writer, the best known agent for the treatment of gastric ulcer. It is also one of the best remedies for the treatment of the stomach, catarrh of chronic alcoholism, and for chronic gastric catarrh from other causes. It is excellent for atonic dyspepsia, and for acid dyspepsia. The writer has seen very gratifying results from its use in these distressing maladies.

In catarrhal and other stomachic diseases except gastric ulcer, the remedy is best administered in one or two teaspoonfuls in a wine-glassful of water immediately after meals. In the case of gastric ulcer the dose and dilution should be the same, but it is better to give it when the stomach is empty.

Glycozone has an excellent effect when used internally in cases of diphtheria. For this purpose a tablespoonful of glycozone is given in a wineglassful of water every three hours. As it is perfectly harmless it may be used without apprehension. The following treatment is excellent in cases of membranous croup: The nose, throat, mouth, pharynx and larynx should be sprayed copiously every two hours or so with a mixture of one ounce of Marchand's peroxide of hydrogen (medicinal), with four to six ounces of water.

The membranes are readily destroyed, and by using this remedy freely, their reproduction is prevented. Then one teaspoonful of glycozone, diluted in a wineglassful of water, administered three times a day, prevents any disturbance of the stomach and regulates the bowels.

Remarkable benefit may be derived in the treatment of diseased conditions (ulceration and chronic inflammation) of the rectum and lower gut, by enemata containing glycozone and for this purpose nothing excels the following formula:

Glycozone, 1 ounce.

Water, lukewarm, 12 ounces.

This should be mixed immediately before using and administered with a hard rubber syringe once daily. It is frequently desirable to use a smaller amount than the above mixture. The proportions 1 to 12, however should be maintained. In cases of fistula-in-ano and of rectal ulcerations low down, an ounce

of lukewarm water containing a drachm of glycozone administered once or twice daily soon effects good and in cases of ulcer, pure and simple, may be expected to radically cure the diseased conditions.

External Uses.—After the cleansing of any diseased or suppurating surface by peroxide of hydrogen (medicinal), the application of glycozone stimulates healthly action and hastens the cure. For this purpose it has no superior in the entire range of therapeutics. It tends to check the discharge of irritating unwholesome secretions and to prevent the infection of the sore by pathogenic organisms. Its action in this respect is explained by the fact that it is both powerfully antiseptic and stimulant.

EDITORIAL.

The Practical Value of the Microscope.—To pharmacists its worth is constantly increasing as physicians learn the value of microscopical diagnosis and druggists become competent to handle it. The editor of a western periodical relates how while in Hot Springs, Ark., he recently saw a very poor microscope that was bought at a high price but had paid for itself several times over. What is more the man who uses it knows but little of the subject. If it is a paying investment under such poor environments what would be the worth of a good serviceable microscope in the hands of one fully competent to manipulate it? He says; "We hope that more of our readers will answer this interrogation by a practical demonstration of the question. The muslin drug stores and saloon pharmacies may cut prices on patent medicines and sell medicines at cost but they will not invest in microscopes."

MICROSCOPICAL APPARATUS.

Construction fo Microscope Stands.—Mr. W. C. Davis, of the Manse, Prestonville, Brighton, claims patent rights on the construction of microscopes with the upright standards sloped backwards (towards the user of the microscope). Another feature of the instrument consists in the adaptation of a curved portion to the supporting hinge of the tube, whereby a convenient

rest for the finger while adjusting the position of the tube is provided. Mr. Davis also describes a plan for reducing the bearing surface of the sliding tube to a pair of narrow strips parallel with the axis thereof. He is thus enabled to lacquer a very considerable portion of the sliding tube, where it is exposed to view, and materially improve the appearance of the instrument. There is, perhaps more that is of a patentable character in this last proposition of Mr. Davis' than in the others—but we do not like it. Admitting that it is an important object, in connection with elaborately finished instruments, to get rid of the appearance which is apt to characterize unlacquered brass, we do not think that the wholesale reduction of bearing surface of the slide is mechanically admissible. As we have said before, and hope to show more fully in due course, the parallel motion linkage is better adapted than the slide for use in high-class philosophical instruments. Slides are very well for rough purposes and machine tools of precision, but they are both unslightly and unnecessary for adjusting purposes in ornamental microscopes and other such apparatus.—*The Optician.*

MICROSCOPICAL MANIPULATION.

Simplification of Laboratory Methods.—Dr. William C. Krauss, Professor of Pathology in the Medical Department of Niagara University, Buffalo, N. Y., refers to the time and annoyance of labelling the bottles containing specimens, to the large number of bottles necessary to keep the various tissues separate and the expense incurred in conducting a laboratory on such a plan. He proposes the following method.

The sections of the various tissues to be later on examined, after hardening and dehydration, are cut in small cubes of 1 to 2 c. centimeters. They are then placed in a wide-mouthed bottle having a cork stopper, on the upper side of which is a deep crease. A piece of heavy card board, 1 to 2 centimeters square with the name of the specimen, case, date and reference written upon it, is slipped into the crease just referred to and the bottle contents are thus securely labelled. During the process of embedding, the original card-board label can be transferred from one cork to another until the specimen is securely fastened upon its own cork ready for section cutting. These spe-

cimens may be stored in a large brain jar, the card-board labels fastened to the corks in the manner above referred to. It will thus be seen that from the time the tissue is placed into the hardening fluid until it has been partly cut into sections and again placed into the storage jar, but one label has been used, and really the use of storage bottles has been dispensed with.

This plan is recommended to those microscopists with private laboratories and to those who are obliged to spend little time and less money in their laboratory work.

MEDICAL MICROSCOPY.

A Study of Palsy.—Dr. Wiener, of Mt. Sinai Hospital, New York, reports a case (N. Y.. Med. Jour., July 14) of subacute unilateral bulbar palsy and its autopsy. He presents five photomicrogrophs which show degenerated ganglion cells. The complete brain of a 17 year old boy was put in Muller's fluid and hardened for microscopic examination. The column known as the respiratory bundle was found to be almost completely degenerated. There was a marked degeneration of the nucleus of the hypoglossal nerve on the right side, together with slight degeneration in the adjacent nuclei. In the paper cited he gives an extended explanation of these facts.

BIOLOGICAL NOTES.

The Smallest Known Flowering Plant.—Mr. Thos. Craig writes as follows:

I have to report having recently found on Staten Island, in the Old Town pond, what is said to be the smallest of flowering plants; a full grown specimen only measuring from 1-16 to 1-32 of an inch. I have not been able to ascertain who was the first to see this plant in flower, nor have I been able to see it myself after careful search with the microscope. Probably the specimens I have are past the flowering season. It is evident, however, that it does not depend entirely on its seed for increase. Like *Lemna*, to which it is closely allied, and which is so conspicuous on all our ponds, it propagates by budding, but unlike the other members of the family the bud immediately separates from the mother plant and becomes independent.

I submit a rough drawing to show this peculiar process and, the specimens may be examined under the microscope. According to the authorities there is a cleft on some part of the plant, out of which the young one grows. So far as I could see there is no appearance of a cleft or opening of any kind until the young plant inside has advanced in growth to a considerable size ; then it appears to protrude, gradually increasing and enlarging the opening, until it finally emerges, leaving a vacancy in the mother plant the full size of the young plant. The opening does not seem to close up. What becomes of it remains for further study and examination to disclose.

Unfortunately there is very little literature on the subject. Only two species are described in Gray's Manual, viz.: *W. Columbiana*, Karst, and *W. Braziliensis*, Weddell—the latter differing from the former in having numerous brown spots over it.

Whether this specimen is *Columbiana* or *Braziliensis* I cannot positively say, but I think it is *Columbiana*. It is, however, plentifully covered with brown spots, but these are the points of attachment of a young alga which is growing on the plant, giving it, with transmitted light, under the microscope, a hairy appearance. Where the alga has dropped or has been rubbed off, a bright reddish brown disk remains. Anyone familiar with the growth of such algæ as attach themselves to objects in the water, will recognize by the appearance which these brown spots have that they are the points of attachment of an alga, this plant has no root and it is very loosely cellular and light green in color.

Dr. Britton informs me that *Wolffia* is recorded from Closter, Bergen Co., and Kaighn's Point, Camden, N. J., and Orange Co., N. Y., and we are now enabled to record it from Staten Island.

MICROSCOPICAL SOCIETIES.

Lincoln Microscope Club, Roscoe Pound, Secretary.

September 26.—This was the first regular meeting after the summer vacation and was unusually well attended. A great deal of interest was shown.

Dr. Bessey exhibited a new Reinhold Giltay Microtome, belonging to the University and explained its construction and advantages.

Mrs. Wade exhibited an old microscope which she had known

for more than forty years, and knew to be much older than that. The contrast it presented to modern instruments, was the subject of no little comment by all.

Among other exhibits, Prof. Seawell showed a section of the eyes of **Pecten**. The next regular meeting will be held October 31st.

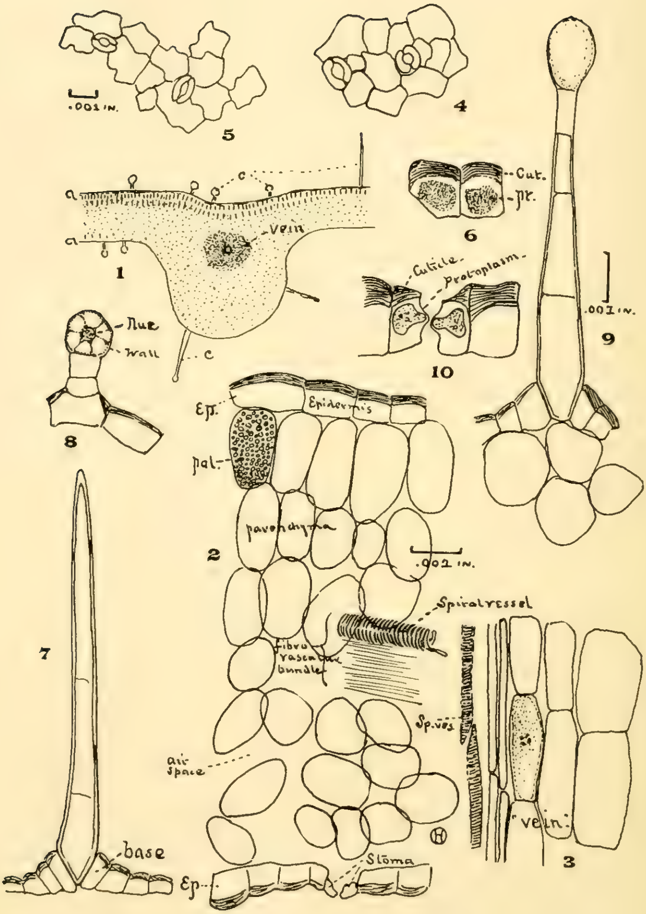
MICROSCOPICAL NOTES.

Microscopical Praxis.—This is the name of Dr. Stokes' new book, just issued. It will be of much assistance to the novice but he who would go extensively into such matters will take up Carpenter or Gage after mastering the outlines here presented, and some may be induced to do so by the pleasing introduction here acquired.

Inoculation of Warts.—It has been found that warts may be produced by inoculation. The microbes inoculated in the epidermic layer multiply slowly, two months having elapsed in some cases. Cultures of wart microbes on agar-agar give greenish-yellow masses. With these cultures inoculation resulted in 15 days in watery excrescences resembling warts. There is no doubt that children with warts should be watched carefully to prevent their contact with other children especially at school.

Primary Dissecting Microscope.—Manufactured by the Bausch & Lomb Optical Co., Rochester, N. Y. This Microscope was designed by Prof. C. R. Barnes of the University of Wisconsin and has been in use in their and other laboratories for several years with very satisfactory results. It is a most effective and low priced dissecting instrument.

The body is a solid block of wood so shaped that the sides serve as hand rests. The advantage of this is that the block practically forms a part of the table on which it rests and is then very steady to work upon. Mirror and movable glass stage are provided for in a very simple manner. The lenses are carried in an arm, the post of which slides in a metal sleeve, thus allowing the entire stage to be covered and giving sufficient rays and accuracy of focus. A square plate black on one side and white on the other is arranged to slip under the stage for dark or white ground. A groove on the lower side of the block receives the plate when not in use.



HISTOLOGY OF THE GERANIUM LEAF.

THE AMERICAN
MONTHLY
MICROSCOPICAL JOURNAL.

VOL. XV. NOVEMBER, 1894. NO. 11.

The Geranium Leaf as a Cell Aggregate.

BY HENRY L. OSBORN,

ST. PAUL, MINN.

WITH FRONTISPIECE.

All the higher or flowering plants, in common with all living things whether plant or animal, are descended directly or indirectly from a single ultimate cell—the "egg-cell." The history of the plant as an individual is the history of this cell, from the time of its fertilization in the maternal ovule by the paternal pollen tube to the time when the plant in its turn produces and matures egg-cells and sends them out to play their part in the cycle of life. The body of a mature plant is the sum of the cells descended from the parental egg-cell in studying which structure we are viewing the vast army of progeny living or dead of the primary egg. In making a histological study of any organism as complex as the geranium leaf, it is very helpful to keep this fact constantly before our minds and to raise the question in examining cells of each kind, what has been their origin and to what other cells are these most closely related ?

Cells in the immature portions of plants are soft walled and delicate. If they should remain so, it is manifest that the plant body could not attain any considerable size because of the weight of the upper layers of cells on those beneath, and that the plant could not live in the air because they would be dried by evaporation and thereby killed. The problem, therefore, for a plant living like the geranium would be to obtain, in some way,

a water-proof outer covering to prevent the loss of the water of the cells and to obtain some form of supporting material to enable it to spread out in the air a considerable surface, and yet one strong enough for support under all contingencies. The requirements of this problem are variously met in all plant leaves, in each kind in a slightly different way. Our present study is designed to show how the problem is solved in the case of the geranium.

The leaf of the geranium is long-petioled, palmately-netted veined, cordate at the base, crenately rounded and lobed, its surface pubescent. An examination shows that the strong and flexible material of the stem is continued out onto the broad blade in the form of several large and innumerable smaller lines called "veins." These run to every part of the structure and form a frame-work for the support of the softer material between. At the margin, minute divisions of the veins can be traced to the lobes and still finer ones to their subdivisions. If the veins of the leaf were to be preserved and the rest removed, the shape of the organ would not be altered in any respect. Besides the supporting framework, the surface of the leaf presents a finely hairy appearance, almost deserving the name "velvety." This is true of both the upper and the under surface and also of the petiole or stem of the leaf. There is a difference in the shade of the two sides of the leaf, the under being somewhat lighter than the upper surface.

After determining these facts, one has a suggestion for studies in the structure of the leaf. He will need to find out what is the cause of the difference in color of the two sides, also how the hairs are made and the structure of the veins. All these facts cannot be ascertained in the same way, but is quite obvious that a good deal ought to be shown by an end view or section of the leaf. Such a view is shown in figure 1. This is made by cutting off

thin pieces crossing a vein and shows the thickening where the vein lies in the center and the thinner blades on each side. A very good way to cut such a "section" as this is to place a small piece of leaf between two pieces of elder-pith and with a sharp scalpel to slice down, cutting as thin as possible. The slices should be floated off into a shallow glass of water and then the thinnest of them floated onto a slide in water and covered for examination.

Figure 1, which is somewhat diagrammatic, was made from such a section. It shows a number of structures which help us to solve the problem of the leaf. We see a thin skin (*a*) covering the central mass of material, and this we suspect of being the stuff which keeps the water of the leaf from instant evaporation in spite of the fact that it is spread out in so thin a layer and exposed to air on both sides. Then we see, in the center, a mass of denser material (*b*) which we recognize as the cut end of the vein, and we see that it is but a small portion of the entire amount of leaf material by which it is surrounded. Then, too, on the surface at various places we see minute projecting bodies which we doubtless recognize as the parts causing the velvety surface. They are called "hairs" and are of three forms. The whole general thickness of the leaf is about one one-hundredth of an inch.

If the section is thin enough, the high power will present a view somewhat like figure 2, which is magnified not far from four hundred diameters. Here two entirely distinct types of cells come to view. On the surface are long and narrow cells (*ep*), whose outer side is very thick and dense. These are "epidermis." They are not green as are the cells in the interior, but are colorless. They are apparently empty, but if a section be properly treated for shrinking the protoplasm of these cells, a layer closely lining the wall of the cell will be discovered as seen in figure 6 (*pr*). In the section beneath the

epidermis there are oval cells with thin walls and filled with numerous very minute green spots or "Chlorophyll-grains." These cells are definite in their arrangement, those of the upper side being tall and vertical to the surface and closely laid. They are called "palisade cells." On the lower side, on the other hand, the cells are loosely laid. There are considerable spaces among the cells into which the gases of the air may pass. This compactness of the arrangement of the upper side is the explanation of the deeper color of that side. Besides these large oval cells, there is in the center—in some places, not in all—a portion of the vein-system. This runs among the cells and supplies the support needed because of their extreme delicacy. It is among these cells of the leaf, in the spaces between them, that the mysterious chemical process of decomposing carbonic acid gas and building up from its carbon such complex products as sugar is conducted. Such being the case, it is at once apparent that the more of leaf, the more chemical reaction. And with epidermis, with its impenetrable outer wall and supporting tissue prevading the entire organ, almost unlimited extent of leaf is possible. The details of the structure of the supporting tissue cannot be undertaken here, but a word about it will be in place. The cells of which it is composed are apparently spirally-wound tubes. In reality, however, they are not strictly spirally-wound tubes, but tubes whose wall is thickened in a spiral line. This gives strength to the tube and at the same time flexibility, enabling it to bend through a considerable distance without breaking short off. That the tubes are cells is shown in figure 3, where the ends of two adjoining spiral cells are shown.

The epidermis is more than a layer of cells with a thickened outer wall. In addition to this, the usual type of cell, there are the various sorts of hairs. Taking plants in general, the number of different kinds of hairs

formed of modified epidermal cells is very great. In the geranium leaf there are three distinct types. One of these (fig. 7) is the sharp-pointed, elongate hair. This presents a number of minor but interesting details. First, it is composed of several cells whose transverse walls still remain. These, on their outer side, are thickened and so matched as to form a single, continuous, strong wall. If these hairs had no thicker walls than the green cells they would break too easily to be of any use in such exposed situations as this outer surface. Not only are they protected thus, but they are propped up at their base by a sort of mound of ordinary epidermal cells (see fig. 7) and are so inserted among them that they are not in danger of being pushed down into the layers beneath.

The second type of hair is the glandular hair of which a specimen is shown in figure 8. Here a couple of smaller cells hold aloft a larger spherical cell, in whose center iodine demonstrates an evident nucleus and strands of protoplasm reaching out to a lining of the same material. A third type of hair is an elongate hair bearing a globular cell perhaps gladular on the summit.

But the most remarkable structure in the epidermis is the stoma or breathing pore, of which great numbers occur on each side. Figures 4 and 5 are surface views made by scraping away the green cells and mounting in water a scrap of the epidermis only. Here the ordinary epidermal cells are seen forming a complete mosaic with no breaks excepting where minute oval bodies of certain constant form appear. These consist of two hemiovals with a very small opening between them. Sections vertical to the surface may pass through the stomata as shown in figure 2. Figure 10 is a view of a section through a stoma of another plant (Hyacinthus) copied from Sach's Text-Book of Botany. It will be seen from these views that the stomata are really openings which

lead to the spaces among the cells of the leaf and thus permit gases to enter or leave. The number of stomata is very great indeed, and they are very essential to the operations of the organ. But the question at once arises, how does the evaporation of water fail to take place when the surface of the leaf is so open as this proves it to be? In figure 10 the cells of the stoma are shown to contain a central protoplasmic material which it is supposed has the function of swelling or contracting, and thereby of closing or opening the entrance to the chemical laboratory of the leaf. These guard-cells, as they are called, by closing in times of atmospheric dryness prevent evaporation, and in more favorable times permit the equally necessary passage of gases.

If a leaf were to be examined while it is still very young, it would be composed of nearly uniform cells throughout. As time elapses, the fibrovascular tissue would appear developed out of some of the at first "indifferent" cells. Later, the chlorophyll grains or green spots in the central cells would make the leaf greenish in color because of their presence; and the surface cells, at first alike, would undergo differentiations, and the stomata and the various hairs be formed from the ordinary epidermal cells.

The leaf is now seen to be made of a very large number of thin-walled protoplasmic cells which contain chlorophyll. Their total number in the entire leaf is verp great and the whole number in an entire plant would almost defy numerical statement. The advantage to the plant of such vast numbers of these cells is seen when we reflect that it is only such cells as these that are able to construct the substances which compose the body of the plant, and that therefore these are the workers which make the building. Thus the size attainable depenbs on the numder of parenchyma cells. In a plant living in the water, there would be hardly any limit to

the size which might be attained by the mere increase of cells of this description; at tthwa tpbnilcuheampts life on land must be able to protect these worker cells or be destroyed. It is thus very clear that noly plants with support and protection for the delicate worker-cells could exist on land, and such only could propagate. There are some plants which live on land, in a certain sense. They lie prone on rocks and are found in damp or wet places and yet have little besides the mass of worker-cells. Such are insignificant and lowly forms mostly unknown beyond the acquaintance of the botanist; but trees, and shrubs, and flowering herbs, all owe their power to live the life they do to the presence of epidermal structures and to the supporting fibro-vascular tissue.

HAMLINE BIOLOGICAL LABORATORY, September 20, 1894.

EXPLANATION OF THE FRONTISPIECE.

These figures are drawn from camera lucida tracings. The accompanying scale of one-thousandths of an inch permits direct measurement of the cells.

Fig. 1. Cross section of leaf passing through a vein (partly diagrammatic).

Fig. 2. Small portion of figure 1 magnified 260 diameters. *Ep*, epidermis. The upper side is up. *Pal*, palisade cells on the lower sive the section passes through stoma. A small portion of a fibro-vascular bundle is shown.

Fig. 3. A portion of the fibro-vascular bundle seen lengthwise showing the junction of two spiral-cells, and the shrunken protoplasm in one of the parenchyma cells.

Fig. 4. Surface view of the cells of the epidermis of the upper side, showing two of the stomata.

Fig. 5. Similar view of the under side, epidermis and stomata.

Fig. 6. Epidermis cells in section after treatment with dilute acid to shrink and demonstrate the protoplasm; *cut* the greatly thickened external wall of the cell; *pp*, the protoplasm.

Fig. 7. One of the hairs showing the wedged-form base and mound of ordinary epidermal cells around it.

Fig. 8. A glandular hair in the outermost cell, a nucleus and vacuolated protoplasm.

Fig. 9. An elongate glandular hair, intermediate type between the other two.

Fig. 10 View of the stoma guard-cells showing the protoplasm *in situ*. Copied from Sach's Text-book.

The Fruits of the Order Ranunculaceæ.

BY KARL M. WIEGAND,

CORNELL UNIVERSITY, ITHACA, N. Y.

An abstract of a paper read before the American Microscopical Society, August, 1894.

The importance of characters drawn from the fruit in the limitation of the larger groups has not been fully recognized. Only the gross anatomy or external features have been used, while the minute structure of the seed remains yet obscure in most of our natural orders.

Actual variation in organisms is probably augmented by changes in environment and modified by natural selection. It follows, therefore, that those parts farthest removed from external influences are the most constant, because they are not required to adapt themselves to any new conditions, and they have long ago become almost as well fitted to perform their function as possible. Moreover, the tendency is when organs have remained unvaried for a long time to loose the power to vary. It is assumed that the function of the organ is also constant, which is not always the case.

The features of a plant most subject to variation through changes in environment are habit, foliage, and parts of the flower. The latter should include the floral envelopes, surface of the seed, and everything having directly to do with pollination and dissemination. The two most constant series of organs in a plant seem to be, first; those connected with nutrition including the internal anatomy, and subject to changes in the soil and atmosphere. Secondly, those organs connected directly with reproduction, especially the internal parts of the seed. These are little affected by environment, and are practically constant if reproduction itself is constant. But there is no essential difference between the way in which new plants are produced in the Polypetalæ and Monocotyledons. Practically the same method is used

by the lower plants in their sexual methods. This shows that the process is practically constant. The embryo is perhaps the most variable of the internal parts of the seed because it comes in contact with the environment after germination, and on the other hand the seed coats are the most constant. Characters drawn from the internal anatomy of the seed should then be those most suitable for the separation of the larger groups of the Phanerogams.

For the purpose of classification, the first groups into which plants are divided must be general, comprising a large number of forms. The distinguishing characters must, therefore, be common to a large number of species. According to our modern ideas of evolution, these must have existed in the progenator of the group, and have remained nearly unchanged in all of its descendents up to the present time. In species and genera, less general groups, the characters may be drawn from parts less constant. The value of a character then depends on its susceptibility to variation.

This idea of the value of the internal features of the reproductive organs in the characterization of larger groups is not entirely new. It has already been acknowledged by most animal embryologists, and in botany the two larger groups of the Phanerogams are separated by features of the embryo.

From this standpoint the present discussion of the fruit of the Ranunculaceæ was prepared.

The fruits of the Ranunculaceæ are mostly either dehiscent several seeded pods or achenes. The achene seems to be the more recent production since the line of dehiscence is still plainly visible between the placental strands of most species, and then too the peculiar structure of the achene with suspended ovules in the genus Anemone lead to this conclusion. Indehiscent fruits are an advantage to the plant only when one seeded, since if

several seeds were bound up together their chances of survival on germination would be decreased. The pods dehisce the whole length of the placenta; hence when this is only at the top of the pod they open at this point. The other form of fruit occurring in the order is the berry found in Actæa and Hydrastis.

The seeds of the Ranunculaceæ are completely anatropus in almost every species. There is no more constant feature in the whole order than this. In general outline they are ovoid with the smaller end toward the hilum, varying somewhat from this only in Caltha, Coptis, and Actæa. The raphe often forms a ring on the side of the seed. This form of the seed probably aids it materially in becoming planted in the ground. Since the small end would be buried first, and this is the end which contains the embryo, with the hypocotyl down, the seed would be in the best possible position for germination. In Caltha palustris the air space at the upper end serves the same purpose as the enlargement in other species.

The embryo represents the extreme minute form of the albuminous type. In many cases it is but little removed from fertilization, and with little or no differentiation into cotyledons and hypocotyl. In general, the smallest and least differentiated are found among the pod forms. The largest and highest developed embryo was found in Delphinium and this also was the only one that showed an indication of the plumule. In most of the larger embryos a well differentiated central cylinder occurs, and branches of this can be seen in the cotyledons of several species. In some genera a root cap can be distinguished. The tissue of the whole embryo is of small cells with large nuclei arranged in vertical rows.

Two seed coats can be distinguished in every genus throughout the order. This result contradicts Engler and Prantl and other authors who say some genera have only one coat. It would seem scarcely probable that a

seed coat well developed in one genus of the order should be entirely absent in another genus especially when we consider the slight variability of these structures.

The inner coat consists normally of two parts; an outer layer of thin parenchyma, and an inner layer with cells regular in outline, thick walled, often striated on the inner side, and thin walled toward the surface. Its color is pale yellow or dark brown. The striations often pass into the cell cavity as ascicular projections. Sometimes, however, the cells are so compressed as to be scarcely visible. This layer is always thickened at the micropyle, but in some cases more than in others. In all pod forms except Paeonia it is elongated into a cylinder passing in most cases to the surface of the seed. This I have termed the collar of the micropyle. In the center of this tissue there is generally a trace of the former tubular canal of the micropyle. This is more distinct in the more generalized forms.

The outer seed coat is often much modified to serve as a protective organ, especially in the pod forms, and consists of two layers. In the pod forms the outer layer consists of one row of latterly compressed cells with thick walls. Their cavities often contain dark coloring matter, and their outer walls form papillæ on the surface. In the achene forms the cells become oblong, rectangular, or irregular in outline, and are so thin walled as to be scarcely distinguishable from the next layer. In all pod forms the cells bend in around the collar of the micropyle and thus line the orifice. In Paeonia, at maturity, the layer is continuous over the micropyle. The inner layer is composed entirely of parenchyma containing dried protoplasmic bodies and rarely nuclei. It is thickened in the region of the raphe the vascular strand of which always passes through its tissue.

The carpels of the Ranunculaceæ are either smooth

rugose, veiny, longitudinally ribbed, or hairy. On the carpels of the achene forms are special provisions promoting dissemination and planting. This we should expect since the carpels always invest the seed and the achene forms are probably the more specialized. This feature is seen in the long hairs of Clematis and Anemone.

There are two distinct layers in the carpellary walls; first, the inner epidermal layer lining the cavity of the ovary; second, the remaining parenchyma outside. The cells of the inner layer are always elongated, mostly square in cross section, and generally thick walled. In most genera they appear like wood cells. This is the layer which has been especially strengthened for protection in the achenes, and is comparatively thin in all pod genera.

The outer layer is several cells thick and composed of parenchyma, except in Ranunculus where it is thick walled. In Actaea and Hydrastis it becomes fleshy. In some of the achenes the cells of the outer layer are slightly elongated and overlap like shingles on a roof, the serration thus caused pointing toward the base of the carpels. The hairs arising between the cells necessarily point downward at first, but immediately make a turn on reaching the surface and point toward the style.

In most of the pod forms the inner cells and ribs of the carpels run horizontally, except near the base where they are longitudinal. In the majority of the achenes they are entirely longitudinal. In the Anemone group the number of ovules has been reduced to one, while the cavity has elongated below and contracted above, leaving the placenta at the top of the cavity and the cells and veins longitudinal. In Ranunculus the ovules are reduced to one, while the elongation of the cavity, if any, has been upward.

The manner in which material was prepared for the

study was as follows: The fruits when fresh were placed immediately in 60 per cent alcohol, or if old and dry were first soaked for a few hours in water. The softened seeds were then dehydrated with 95 per cent alcohol by means of a Schultz's dehydrating apparatus (see Thomas, Collodion Method in Botany, Proc. Amer. Mic. Soc. 1890) for about seven days. The material was now placed in 2 per cent collodion (2 grams. gun-cotton, 50cc. 95 per cent alcohol, 50cc. ether) where it remained abont four days. It was then transferred to 5 per cent collodion. Two days later this collodion with the seeds was poured into shallow paper boxes and hardened in alcohol. Square blocks containing the seed could then be cut out and placed in any desired position on cork, and fixed in place by the addition of a few more drops of collodion.

The sections were all made on a sliding microtome with a rather narrow and thick bladed knife. They were stained with hematoxylin and mounted in series in Canada balsam. Sections were taken in two vertical planes and transversly.

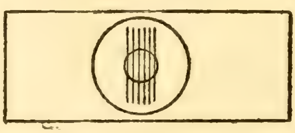
A Marking Apparatus for Indicating the Position of Objects or Parts of Objects in Microscopical Preparations.

By S. H. GAGE,
ITHACA, N. Y.

Doubtless every one who works much with the microscope has felt the need of some device to enable him quickly to find what he knows to be under the cover-glass in a given preparation. This applies of course with especial force to the higher objectives where the field is so small. Various devices like the Maltwood finder etc., have been invented for this purpose. Special verniers have been added to mechanical stages for the same purpose. The perfect mechanical stage with verniers is all

that could be desired, but these cannot be supplied with every laboratory microscope and neither can the stops for the Maltwood finder be used in all cases. To attain the object of marking preparations and at the same time leaving the microscope and the preparation in as simple a condition as though no special marks were used was attained by Dr. May of Philadelphia, and later by Winkel of Goettingen, by means of an objetive like apparatus in which a diamond point might be made more or less eccentric, depending on the circle desired, and then by rotating the carrier of the diamond a slight scratch was made on the cover enclosing the object. The difficulty here is that the line must be so fine that it is difficult to find the circle in the first place and in the second place homogeneous objectives cannot be used as the fluid would obliterate the diamond line unless they were filled with plumbago and protected in some way.

To overcome these difficulties and mark the specimens easily and so plainly that there was no trouble in finding the right field, the writer for many years, made an ink mark near the edge of the field with a fine pen, then placed the slide on the turn table and made a delicate circle of colored shellac or other cement around the place indicated. This answered fairly well, but to be able to have a marker on the nose-piece and whenever any object was found that showed a special structure well seemed so desirable that the device of May & Winkel was modified by using a small brush. This brush was attached to a piece that could be made eccentric, then this to another, rotating on a central axis which was screwed into a piece with Society Screw, which in turn could be attached to the nose-piece. With this simple apparatus every object showing a particular structure well can be marked in half



a minute, the brush turned aside and the study of the specimen continued.

It has been found of the greatest convenience for demonstrating preparations. The right thing can be found in a very short time. For reference preparations for students it has also proved a great assistance. The teacher can also feel confident that the student will find and study the typical part of the preparation.

The Histological Structure of the Enteron of N. Maculatus.

By B. F. KINGSBURY,

ITHACA, N. Y.

The large-gilled salamander of the lakes and rivers of North America, *Necturus maculatus*, known to fishermen as the "mud-puppy" or "water-dog," presents exceptionally favorable opportunities for the study of the structure and physiology of the digestive tract, surpassing in many respects even the much-used frog. When it is considered that the cells of the stomach epithelium of Necturus are about three times as large as those of the frog or cat, it may be recognized how clearly structure and the changes accompanying functional activity can be observed.

The digestive tract in this animal is quite simple, and consists of a mouth cavity, short, wide, esophagus, a long tubular stomach and an intestine with two or three convolutions opening into a cloaca. A distinction between a small and large intestine such as occurs in the majority of Amphibia is wanting.

The mouth is lined with a stratified epithelium, which is non-ciliated, a condition the reverse of that found in the majority of Amphibia investigated. As far as can be judged, those Amphibia in which cilia are wanting in the mouth are those whose entire life is spent in the water. The epithelium cells here resemble very much those of the epidermis. No salivary glands, or glands

of any kind are present. Mucous is however abundantly secreted by single cells of various shapes and sizes which occur among the other cells in the mouth and esophagus. In most of these respects, however, it does not differ from other Amphibia.

In the stomach, two kinds of glands are found, as is the case with the majority of vertebrates, though their homology with the glands occupying the cardiac and pyloric portions of the stomach in mammals is doubtful.

The cardiac glands are far the more numerous and the arena occupied by them is clearly demarcated from the pyloric region by the greater opacity of the mucosa, due to the granular cells of these glands.

The structure is that characteristic of these cardiac glands in Amphibia. The surface epithelium dips down forming the neck of the gland, below which are several large mucous cells, which are in turn followed by a tubule formed of the secreting cells. In mammals pepsin is now thought to be secreted by the chief cells of the cardiac glands, while the parietal cells produce the hydrochloric acid of the gastric juice. In Amphibia no parietal cells are present and the secreting cells seem to perform both functions. They contain numerous granules which embody the precursor of the pepsin, and their abundance has been shown by Langley in investigation on the frog to depend directly upon the state of digestion. These granules may be clearly seen by hardening the tissue in a 1 per cent aqueous solution of osmic acid, which stains them a light brown. In the pancreas of Necturus also the zymogen granules of trypsin may be beautifully demonstrated.

Leucocytes of white blood corpuscles occur in all portions of the alimentary canal in the connective tissue and in the epithelium, where they are sometimes of enormous size, and greatly distort the epithelium cells surrounding them. In the stomach epithelium they are to be found

constantly, and are often filled with granules presumably fat since they blacken with osmic acid. But not only fat seems to be appropriated by these cells; they often contain densely staining globules and protoplasmic masses and also other smaller leucocytes. This seemed to be more often the case in those found in the intestine and epithelium of the mouth cavity, where they were often quite packed with globules, some of which took a nuclear others a cytoplasmic stain, and were apparently the remains of leucocytes which had been engulfed. In addition to these large leucocytes, to which the name used by Ruffer, macrophages, might be appropriately applied, other leucocytes occurred, both mono and multinuclear forms, and leucocytes with much or scanty protoplasm. Particularly abundant, especially in the subepithelial tissue of the mouth were small leucocytes with granules which were stained a bright red, undoubtedly the eosinophile leucocytes of Ehrlich.

For fixing the tissue of the digestive tract mercuric chloride was found to give excellent results. It was employed as a saturated solution in normal salt solution (water 100 c. c., salt 6-10 grams). The tissue was fixed in this only long enough to enable the fluid to penetrate to all parts, generally one or two hours. The tissue was then washed well in 67 per cent of alcohol, to which a little gum camphor was added to ensure the complete removal of the mercuric chloride. Tincture of iodine may be used as a test for this; which is discolored by mercuric chloride. Picric alcohol also gave excellent results. The formula employed was as follows: 95 per cent of alcohol 250 c. c., water 250 c. c., picric acid crystals, 1 gram. Tissue was hardened in this from 12-24 hours, and afterwards with 67 and 82 per cent of alcohols.

The Ehrlieh-Biondi triple satin was employed with excellent results. The mixture as prepared by Dr. Grubler

of Leipzig (100c. c. saturated aqueous solution of Orange, 20 c. c. saturated aqueous solution of Acid fuchsin, 50 c. c. sat. aq. sol. Methyl Green.) was diluted 60 times and the section allowed to stain 2 or 3 hours, then washed and dehydrated with 95 per cent of alcohol, cleared in zylol or carbol-xylol (Carbolic acid crystals 1 part, xylol 3 parts) and mounted in xylol balsam. It is necessary that the alcohol with which the stain is washed out, be neutral as, if acid, the methyl green is washed out, and if alkaline the fuchsin is faded in the result. Tissue hardened in mercuric chloride solution, or with picric alcohol may be stained with this with good results. It was employed however only with tissue cut in paraffine, since the readiness with which collodion was stained by the methyl green rendered it less satisfactory when that method of imbedding was employed.

Microscopical Technique Applied to Histology.—VIII.

[FROM THE FRENCH OF RENÉ BONEVAL.]

(*Continued from page 318.*)

THE PERIPHERAL NERVES.

Dissociation.—It is by dissociation that the study of the elements entering into the composition of the nerves should be begun. We select two, the one of nerve fibres with myeline, the other of fibres without myeline, or those of Remack.

The frog's sciatic is formed of a single nervous bundle which bifurcates at the lower extremity. This arrangement renders dissociation especially easy, and it is the nerve to be chosen for the study of isolated nerve tubes. After destroying the frog's spinal chord by the aid of a needle slipped into the spinal canal, skin the animal and lay it on its belly. The femoral triceps above and the semi-membranous below define a space which points out the position of the sciatic nerve. Carefully removing

these two muscles, we see between them the nerve, which we isolate down to the knee joint, on a level with which it bifurcates. Take every precaution to touch the nerve as little as possible, and especially avoid seizing it with the forceps. A blunt probe will serve to raise it. The nerve must be fixed in a state of physiological extension. Ranvier's process being classic it is well to repeat that learned histologist's experience point by point. Take a little stem of wood, a match for instance,. . . .slip it under the nerve and fasten the latter by two ligatures, taking care to put one below the bifurcation. Cut the nerve beyond the ligatures and put it in 1 per cent osmic acid. . . . .As beginners often stretch the nerve too much, the following modification may be used. The nerve being well isolated, with a pipette filled with 1 per cent osmic acid, wet it while it is still in place. A few drops are enough if the pipette be passed along the length of the nerve. In 3 or 4 minutes it may be cut and will not contract; put it in the osmium solution for ½ an hour, wash in a large quantity of water. It is in this bath that we begin the dissociation. Sieze each bifurcating fascicle with fine forceps, and tear gently. The nerve being divided into two fascicles, repeat the operation upon each until a fascicle is obtained containing only a few nerve tubes. Place on a slide, examine with a low power, and if sufficiently dissociated, add a drop of alum carmine which is to be replaced by glycerine when the nuclei are stained.

Fibres without myeline, or Remack's fibres, exist in considerable number in the pneumogastric nerve. This may be had from the dog, the rabbit, man, etc., but as it is easy to be found in the frog we select that animal. . . . A frog, immobilised by piercing the spinal chord, is put on its back, the sternum cut in the median line and the two parts of the thoracic walls are separated so as to expose the pericardium and the lungs; introduce a strong

glass rod into the æsophagus. We then see (1) the aorta, ...against the cartilaginous extremities of the posterior horns of the hyoid bone ; (2)....muscular fibres extending from these horns to the occipital region ;....the lowest of the muscular bundles is the *satillite* of the pneumogastric which extends along its lower border ;....a nerve extended on a match is placed in 1 per cent osmic acid for 24 hours. Wash for 12 hours, harden in alcohol and gum; section and stain in picro-carmine for 12 hours, wash, mount in water which is to be replaced by glycerine.

To be continued.

Keys to the Genera of Pediculidæ and Mallophagidæ.

BY HERBERT OSBORN,

AMES, IOWA.

The following tables which are in part adapted from the tables given in Piagets *Les Pediculines* and other works will enable one to determine the genus of the parasitic insects infesting birds and animals.

HEMIPTERA.

SUB ORDER PARASITA.

PEDICULIDÆ.

Wingless suctorial insects with clasping tarsi Parasitic upon Mammalia.

KEY TO GENERA.

A Thorax wider than abdomen. Body wider than long. Abdomen with 6 apparent segments. *Phthirius*

AA Thorax narrower than abdomen. Body longer than wide.

a Abdomen with 7-8 segments (on Primates).

b Antennae five jointed on (Homo). *Pediculus*

bb Antennae three-jointed (on Simiadæ). *Pedicinus*

aa Abdomen with 8-9 segments (on lower Mammalia).

b Antennae five-jointed.

c Head normal, antennae slender. *Haematopinus*

cc Head produced, tubular in front, antennae enlarged in middle. Infesting elephant. *Haematomycus**

bb Antennae four-jointed *Echinophthirius**

bbb Antennae three-jointed. *Haematopinoides*

*Not yet recorded for North America.

MALLOPHAGA.

MALLOPHAGIDÆ.

PHILOPTERIDÆ.

A Antennae of five-joints.
  a Antennae similar in two sexes.
    b Forehead truncate, emarginate, more often rounded, not crenulate, last segment round or notched.
      c Large broad trabeculae large, free. *Docophorus*
      cc Trabeculae small, fixed. *Nirmus*
    bb Forehead profoundly crenulate. *Akidoproctus*
  aa Antennae differing in two sexes:
    b Wide, body rounded, or oval elongate. Temporal lobes usually angular, last segment of male rounded or in some cases with two points.
      c First joint antennae male large, sometimes with an appendage, third joint always with an appendage. *Goniodes*
      cc First joint large but without appendage. Last segment of abdomen always rounded. *Goniocotes*
    bb Narrow, body elongate. Sides almost parallel. Last segment male notched.
      c Third joint male antennae without appendage. Temporal band forming a ridge, behind eye; a second abdominal band parallel to border. *Ornithobius*
      cc Third joint male antennae with appendage, no ridge to temporal band or second abdominal band.
        d Antennae and feet well developed. A semicircular fossette in front of mandibles. *Lipeurus*
        Antennae and feet short, in place of the fossette a rainure or depression extending to anterior border. *Oncophorus*
AA Antennae of three-joints. *Trichodectes*

LIOTHEIDÆ.

A Tarsus with two claws, (infesting birds).
  a Head with a deep orbital sinus.
    b Head rounded, without lateral swelling, the antennae passing the margin. *Colpocephalum*
    bb Head with strong lateral swellings.
      c Antennae passing margin of head. Temporal lobes protuberant with right angle. Eye very large and simple. *Boopia**
      cc Antennae hidden. Temporal lobes rounded or slightly angular. Eye parted by a spot.

d Meso-thorax separated from Meta-thorax by a suture. *Trinoton*

dd Meso-thorax united to Meta-thorax without suture. *Laemabothrium*

aa No orbital sinus or if present shallow or covered by an expansion of upper part of head.

b Sides of head straight or a little concave. Forehead with two palettes. *Physostomum*

bb The sides of the head sinuous without palettes.

c Body very broad. Meta-thorax shorter than pro-thorax. *Eureum**

cc Body elongate. Prothorax shorter than meso-thorax and meta-thorax together.

d Orbital sinus occupied by a strong swelling, the bands of the sternum forming a quadrilatera without median spots. *Nitzschia*

dd Orbital sinus without swelling, slightly indicated or wanting entirely. Sternum with median spots. *Menopon*

AA Tarsus with a single claw, (infesting guinea pig). *Gyropus*

*Not yet recorded for North America.

To Mount Certain Salts.

By NO SIG.

PARIS.

DI-ACETATE OF COPPER.

Di-acetate of copper is a very good example of a dichroic crystal. It shows a strong blue color in one direction and a bright green in the other. To see this effect, it is only necessary to use the polarizer, the analytic effect of the salt supplying the place of the analyser.

This salt should be allowed to evaporate spontaneously, and the slide must be thoroughly dried before the salt is mounted.

The crystals will stand the heat of the hot plate without any danger of their being spoiled; so that they could be mounted in balsam with great facility, but unfortunately when mounted in balsam, although the crysals are pretty distinct, they are surrounded with a halo

of color as though they were partially discolored, so that it makes a much brighter slide when mounted in castor-oil. In mounting, let the first drop of oil run to waste, and drop sufficient oil on to well cover the preparation. As the crystals are far from flat, it will take several drops of the oil to fill the space occupied by the cover-glass. Before covering with the glass, put the preparation away for a day under a bell-glass to allow the air-bubbles to escape; and, before mounting look at the slide under the microscope to make sure that is worth being completed.

When you have some thickness of oil contained between the slide and the cover, it is necessary to be very careful not to remove any of the oil by the brush in cleaning the edges. A good plan is to tack one side with a small drop of lac-varnish, and let it dry before proceeding to ring the slide. The cover resting on the tops of the crystals is almost sure to be displaced by the brush, when you try to trim a ring on the turn-table without this precaution. By getting a small spot on one side and drying it, it gives a sufficient hold to the cover to prevent it slipping under the pressure of the varnish brush. This can also be effected by putting a small conical pistol bullet on the glass cover after it is centered on the turn-table, the weight preventing the cover being shifted by the brush.

This makes a very good slide and nearly every one will crystallize uniformly and with well-defined and clear crystals. The crystals are much better if allowed to dry spontaneously than if dried off on the hot-plate; although, of course, the size of the crystals vary according to the heat of weather in which the slide is prepared, in cold weather the crystals being larger.

OXALATE OF CHROMIUM AND POTASH.

This salt gives a very brilliant slide and is also di-

chroic, but not very strongly so. But it is interesting to observe, with the polarizer alone, the color given by the thicker crystals. They are blue in one position and faint red or yellowish according to their thickness when turned half way round. The salt crystallizes best when allowed to dry spontaneously and is best mounted in castor oil. Balsam affects slightly. the brilliancy of the preparation. As some of the crystals are pretty thick, the same precautions are necessary as in mounting the di-acetate of copper.

It may be useful to give the details for preparing plates of the iodo-sulphate of quinine called Herapathite, which can be used to replace a Nicoll's prism or analyzer. This description is taken from Dr. Golding Bird's "Elements of Natural Philosophy," 1860.

To prepare this salt for the formation of polarizing laminæ, the following plan may be adopted :—Dissolve 50 grains of di-sulphate of quinine in two fluid ounces of acetic acid, and two of proof spirit, warmed to 100 degrees F. in a very wide-mouthed flask or glass beaker. Then slowly add 50 drops of a solution of 40 grains of Iodine in an ounce of rectified spirit. Agitate the mixture, and then set it carefully aside for six hours, in an apartment maintained at a temperature of about 50°F. The utmost care must be taken to avoid any motion of the vessel. Indeed, all accidental vibration should be guarded against by suspending the vessel by a string, or by allowing it to rest on a mass of cotton wool.

If, after six hours, the large laminæ of the salt have not been formed, warm the fluid with a spirit lamp, and when it has become clear, add a few drops of the solution of iodine in spirit. The large laminæ form on the top of the fiuid, and should be removed carefully by gliding under one them a circular piece of thin glass.

The specimen should be drained by resting the edge of the glass on a piece of bibulous paper, but it must not

be touched on account of its extreme fragility; if any small crystals adhere to its surface, they must be washed off by pouring over it a few drops of watery solution of iodine. When dry the specimen should be placed for a few minutes under a bell-glass, by the side of a watch-glass, containing a few drops of tincture of iodine; and lastly, a little very-fluid Canada balsam should be dropped on it, and a thin glass cover be applied without heat.

Specimens may thus be obtained of extreme thinness, and half an inch in diameter, or even larger, possessing scarcely the slightest color, and yet completely polarizing transmitted light.

The Maltwood Finder and Similar Devices.

By A. C. STOKES, M. D.,

From his new book "Microscopical Praxis."

Reference has already been made to the difficulty of finding a small object, or some special part of a larger object, with a high-power unless a mechanical stage be used. The field of the high-power objective is so small that the chances of bringing the desired object within its circumference are slight. Usually it is necessary to substitute a low-power lens, bring the specimen within its field, then to re-attach the higher-power objective, in or near whose field the object should be.

On mechanical stages there are commonly engraved two sets of lines about one one-hundredth inch apart, the scales resembling those of a micrometer. These are intended to facilitate the finding of the object the second time, the objective being noted, and the position of the stage recorded as read from the horizontal and the vertical scales on its surface. When the stage is again placed in those positions, the same objective used, and the slide laid on the object-carrier as it was before, the object should then be in the field.

Several devices have been suggested for the convenience of those microscopists that do not possess a mechanical stage. These consist of lines photographed or ruled on a glass slip, their number usually being great and the spaces between them small, each of the latter bearing one or more figures. The best known of these finders is Maltwood's, a glass plate bearing twenty-five hundred squares, so numbered that the position of an object may be recorded by recording the numbers within the space over which it may be when in the field of a certain objective.

The object is brought into the centre of the field of view, when the slide is removed and the Maltwood finder substituted. The numbers on the square now occupying the position previously occupied by the object are noted, and whenever this special square is again brought into the field of that objective, the stage will be in a position to bring at once the desired object into that field.

EDITORIAL.

Sawing Rock Sections.—The Geological Survey in Washington uses a band saw which consists of an endless steel wire $\frac{1}{8}$ inch thick running at a high rate of speed over two fly wheels. Water and emery are fed upon the wire when in motion and the hardest rocks yield to it.

With grinding machines sections one-thousandth of an inch are made and they are of course perfectly translucent, under proper illumination. After one side has been ground flat, a stone is cemented to a piece of glass by means of Canada balsam. A slice can then be sawn off and ground down to within the least possible distance from the glass. It is then removed from the glass by softening the Canada balsam. Mineralogists now regard this method of identifying rocks as favorably as the chemical method in the measurement of crystals.

The Ruby.—A pure, limpid, fiery red piece of corundum is called a ruby. It crystalizes in many different shapes. By submitting it to polarized light its structure is seen to be very

complex. Under the microscope its exterior face is covered with a strange net-work of sculpture, indicating molecular changes. The crystals often have foreign bodies enclosed within their walls—a bubble of gas or a drop of liquid. These microscopic cavities are often very numerous. One which contained carbonic acid was exploded in Edinburgh by being warmed in the mouth.

MICROSCOPICAL APPARATUS.

Substage Condenser.—According to Peragallo, the plane mirror suffices for all objectives of an aperture not exceeding N. A. 0·75—that is, up to about ¼in. objective—and this is true if you have plenty of light, but on dull days a condenser is useful with any objective from ½in. upwards. Above ¼in. a condenser becomes really indispensable, as otherwise you will not get sufficient illumination, especially when using daylight. You will want all the light the condenser will collect on most winter days when working with the 1-12in. and upwards. A condenser also enables one to illuminate with very oblique pencils, which considerably aids the visibility of certain fine surface markings. Also in certain cases you want to remove the morphological details as much as possible, so as to leave a nearly pure color picture, as in bacteriology, and for this purpose a condenser of sufficient aperture is indispensable. I do not think the condenser improves the definition of an inferior objective. A condenser makes the faults of an objective more evident. But with high-power wide-angled objectives all authorities agree that a condenser of some kind is absolutely requisite, and also that there should be some correspondence between the angle of the objective and that of the condenser, though what precisely that correspondence should be authorities do not quite agree. The only utilisable portion of the cone of light from the condenser is the aplanatic portion. This in condenser N. A. 1·2—is about N. A. 0·5, amply sufficient for any objective up to a 1-12in. It is the fashion now amongst the ultra-refined microscopists to speak slightingly of these chromatic condensers of Abbe; but excellent work has been done, and is being done, with them.

Zeiss does not admit that achromatism of the condenser is essential to correct microscopical vision, though better for photo-

graphic purposes; and Naegeli and Schwendener, seem of the same opinion.

For years I used a condenser I made myself. It cost me nothing, as I had the apparatus, but would only have cost me a very few shillings if I had needed to buy it.

A small microscope is sold at about the price of 1s. Standing on three legs, and containing two good large biconvex glasses, the body screws out of the rim of the stand, and if wrapped round with two thicknesses of chamois leather will tightly fit the understage ring tube. I then got two small biconvex glasses out of an old three-power hand magnifier, and cut the horn rims so that they fitted the aperture of the stage tightly, the smaller lens being uppermost. I then arranged the large lenses so as to get a focus on the surface of a thin slide—that is, the smaller lenses being in place—and the thing was complete. It served its purpose perfectly, as far as my requirements were concerned, and gave me sufficient light on the dullest day of winter to use a water-immersion 1-16. The large lenses were kept in position by rings of stiff paste-board; room was left above the uppermost lens so that the diaphragm might just clear. I believe I could do anything with the objectives I employed in the way of resolving markings that they were capable of, though I did not go much in for that perhaps somewhat overdone thing. I could get a very complete removal of the former picture when examining tinted bacteria—my usual work; but, of course, the aperture of my condenser had to be greatly cut down to see podura markings, or blood corpuscles, or histological details, even with a 1-16in. LANCASTRIAN.—*Eng. Mech.*

A New Microscope for Observations at High Temperature.—With a view to examining the transformation of dimorphous substances at temperatures up to 600°, Mr. Von Wyronboff has had constructed a polarization microscope, by Nachet, which is much simpler than the instrument previously employed by Lehmann and others, but which nevertheless, withstands the radiations of the heated objects for a longer period. These advantages are obtained by giving the objective a very long focus. The polarizing nicol is placed at an adequate distance above the stage. The image is obliquely thrown upwards, and the microscope is suitably bent, whereby the observation can be made in the usual position. The object lies on a per-

forated ring-shaped sheet of copper, which can be heated as desired by means of two Bunsen burners. It is also affixed to a special insulating support to prevent any heat being conducted to the body of the microscope. As the object cannot, therefore, be turned or moved in its plane, the entire microscope (with which the polarizer is firmly connected) can be made to revolve in a horizontal semi-circle, and it can also be moved horizontally by means of two guides. The motions provide for the measurement of inclinations due to extinction. The instrument does not exactly measure temperatures, but some thermometric substance. This microscope is in the opinion of the *Zeitschrift fur Instrumèntenkunde*, not suitable for making observations in convergent polarized light.—*The Optician.*

Professor Gage's marking apparatus.—Attention is called espically to the article on pages 334-5. The figure represents a stage micrometer with a delicate ring of cement enclosing the band of lines. This has the advantage of making the observer certain that the lines are somewhere within the circle, and furthermore, by focusing on the edge of the circle first, there is less danger of breaking the micrometer as one knows he is about at the point where the lines should appear.

MICROSCOPICAL MANIPULATION.

Freeborn's Method of Collecting Material—At a meeting of the New York Pathological Society, held September 23, 1891, Dr. G. C. Freeborn made a report on a Method of Rapidly Collecting Deposits for Microscopic Examination, in which he called attention to Litten's report, and himself demonstrated a home-made machine capable of about nine hundred and fifty revolutions per minute. This had answered the purposes for which it was devised admirably. He concluded that "in ordinary urinary analysis not only would this new method enable the microscopical examination to be made about twenty-four hours sooner than formerly, but it would effectually prevent decomposition of casts, which was liable to occur when the urine had to stand for many hours in order that the deposit might settle."

A Transparent Cement.—For cementing purposes, opticians get the oldest Canada balsam they can obtain, and drive

off nearly all the essential oil left in it, by long continued moderate heat; the residual resin is then made slightly less brittle, by dropping into it, when melted, an exceedingly small portion of castor oil; it is easy to add too much of the latter. The object is to get a transparent cement which will neither crack with age nor permit the formation of aborescent markings between the glasses from evaporation of essential oil.—*Photo-American.*

To Prevent Vegetable Sections Turning Dark.—Soak in alcohol to which 2 per cent of hydrochloric acid has been added and let them dry slowly.

Bleaching Leaves.—Chlorinated soda is the best fluid for bleaching vegetable specimens.

BIOLOGICAL NOTES.

Blood Measurements.—In *The Microscope* for May, 1886, was published a paper by Dr. A. Waterhouse giving the results of years of labor in measuring blood corpuscles. He had hoped to be able to distinguish human blood from that of lower animals. As we have frequent calls for that number, which is out of print, we repeat here the gist of all that the article contained. His method, after many experiments with different methods was to use fresh blood spread upon a thin glass cover so thinly that the corpuscles should not touch each other and become disturbed. He warmed it gently so that the blood should dry quickly. A ring was made with cement on a slide and the cover inverted upon it so that the blood would be on the under side of the cover. The ring keeps the blood from touching the slide and forms a closed cell. In it the blood keeps for years.

The measurements were made with a fine 1-16 inch objective and D eye-piece micrometer giving a magnification of about 2,800 diameters. Value of his eye-piece micrometer was determined by a stage micrometer ruled by Prof. W. A. Rogers, to 1-2500 of an inch. The relative length of tube, from eye-piece to objective, was kept the same, and the collar adjustment was placed at the same point of correction.

His largest corpuscles were those of the foetus of grey gopher (1-2,009 in.) and the smallest those of the goat (1-7,031 in.).

Those of man varied all the way from 1-2,812 in. to 1-5,625 in., the latter being those of a man 80 years old.

His list of measurements included the following:

The averages of 9600 measurements of Mammalian blood corpuscles, given in millionths of an inch.

Note.—The corpuscles of each animal mentioned below were measured 200 times for each item given except in the cases of the Terrier Dog, the fœtus of Grey Gopher and the Black Bear.

	Maximum.	Average.	Mimimum.
Grey Gopher	462	325	249
Fœtus of Grey Gopher (100 meas.)	497	429	355
Indian Elephant	426	399	302
Fœtus of Cat	391	333	284
Fœtus of Cat	391	320	284
Gray Rabbit (Lepus Sylvestris)	355	326	284
Gray Rabbit (Lepus Sylvestris)	355	322	284
Gray Rabbit (Lepus Sylvestris)	332	302	213
Woodchuck (Arctomy monax)	391	326	284
Guinea Pig (Cavia cobayia)	355	323	284
So. Amer. Ape	373	325	249
So. Amer. Ape	355	321	249
Terrier Dog (100 meas)	355	316	284
Water Spaniel Dog	355	318	249
Rabbit (Lepus domesticus)	355	317	284
Rabbit (Lepus domesticus)	355	310	284
Newfoundland Dog	355	317	302
Newfoundland Dog	355	315	284
Mongrel Dog	356	314	267
Grey Hound	355	320	267
Setter Dog	355	309	249
Fox Hound	355	295	249
Prairie Dog (Cynomus)	355	289	231
Man	355	328	266
Man	355	328	267
Man	391	323	248
Man	355	320	284
Man	355	319	284
Man 80 years old	320	252	178
Horse	284	231	178
Ox	231	199	160
Sheep	213	179	160
Mouse	284	239	160
Goat	199	165	142
Red Fox	284	253	221
Red Fox	267	242	213

Red Squirrel	320	286	267
Black Bear (100 meas)	320	280	249
Star-nosed Mole	338	270	178
Weasel	338	266	213
Rat	284	265	231
Skunk	284	255	213
Chipmunk	284	260	231
Chipmunk	248	217	151
Ass	320	258	213
Hog	284	255	213
Little brown Bat	284	254	231
Raccoon	284	250	213
Cat	266	222	195
Cat	267	217	178

MEDICAL MICROSCOPY.

Hydrogen Dioxide.—This remarkable liquid which con, tains the greatest percentage of oxygen of any compound known was, for sometime, considered as a mere solution of oxygen in water, and consequently was called oxygenated water. It was afterward obtained free from water and found to be a definite chemical compound of hydrogen and oxygen, and differing from water in containing twice as much oxygen. In this state it is a heavy, oily liquid, readily decomposed at ordinary temperatures- but if heated, with explosive violence, being converted into ord, inary water and oxygen gas. When poured into water it sinks, being nearly half again as heavy as that liquid, but is miscible in all proportions with it. It has a somewhat bitter, astringent taste, and is colorless, transparent and without odor. It bleaches the skin, hair, ivory and destroys organic coloring matter, pus and all organisms with which it comes in contact by liberating oxygen gas in a nascent or active state.

The preparations found in commerce are only solutions of this compound in water, and sold in different degrees of concentration or strength, rated by the number of volumes of oxygen gas they can be made to yield. A fifteen volume solution is one that will give off fifteen volumes of gas from one volume of the solution. A ten volume solution will yield ten pints o- oxygen gas from one pint of the solution, and so on.

These solutions, although more stable than mere concentrated preparations, nevertheless decompose and lose their nascent

oxygen on which its powerful antiseptic powers depend, and consequently we find the commercial brands varying considerably from their reputed strengths. The solution containing the greatest percentage of available oxygen is the preparation known as Marchand's, which, when perfectly fresh, is about a fifteen volume solution.

BACTERIOLOGY.

A Co-operative Bacteriologic Investigation.—The committee on the Pollution of Water Supplies, of the American Public Health Association in its report submitted at the meeting in Montreal, Canada, September 25th last, suggested a co-operative investigation by American bacteriologists into the, at present, confused state of our knowledge on the important subject of the bacteriology of water supplies. The Association approved this suggestion and commended the efforts of the Committee in carrying out the work to the officers of State and municipal boards of health, to the individual members of the Association, and to all persons interested in the purity of water supplies, for such special assistance as they may be able to render. The proposition of the Committee was to the effect that each laboratory willing to co-operate in the work devote itself particularly to the investigation of a single so-called group or species of water bacteria; and that uniform standard culture media be adopted, with the employment of these along the line of a definite systematic scheme of laboratory work. The Committee when its membership is complete, will consist of those participating in the work. In the meantime its organization is in progress.—*Journal American Medical Association.*

The Diphtheria Bacillus.—Strenuous efforts are being made to correctly distinguish the virulent and non virulent diphtheric bacilli. Morphologically they are identical, and find a home in the throat of healthy people as well as those suffering from diphtheria. Bacteria from a healthy mouth and throat are non virulent as a matter of course, while those taken from an infected site naturally convey the poisonous virus.

MICROSCOPICAL NOTES.

Improper Use of The microscope.—The microscope has

proved of inestimable value to medicine. Much of our progress may be attributed to this source. But it has also served to perpetrate some very serious errors. It has one grave fault, you can see in it almost anything that you wish to see. It has but one eye and that is introspective. Moreover, its too exclusive use causes a sort of mental myopia, in which the individual can only see the magnified object before him and note its proper relation to other things.—*Medical Brief.*

NECROLOGY.

A. H. Breckenfeld.—It is with great sorrow that we announce the death of a prominent microscopist and a personal friend. He was a prominent member of the San Francisco society and was recently its president. He was an unusually genial and obliging friend. Every spare hour was spent by him in microscopical study or in roaming the fields and seashore in search of objects. His wife always accompanied him and was his idol. Without her by his side he did not care to live.

The library which he had collected consisted of several hundred valuable books and has been given to the San Francisco society and will be known as the Breckenfeld collection. To these, he added a very handsome monogram slide of arranged diatoms which had been presented to him by the California College of Pharmacy. Of the use to be made of the books, he said: "It is my sincere wish that they may be the means of conferring upon others the same pure and lasting pleasure which I have derived from them myself. Whether this end can be best attained by keeping said collection intact or by distributing it among the members, I leave entirely to the society's good judgment. My own wishes will be gratified if as many as possible are made as happy as possible."

MICROSCOPICAL SOCIETIES.

San Francisco, Cal.

October 3.—Owing to the reception of the Breckenfeld collection and other causes the society has been obliged to enlarge its rooms. At the same time a very wise step has been taken in reducing the entrance fee from $20 to $10. Provision is already

being made to receive the American Association for Advancement of Science and kindred organizations next summer. Great hopes of benefit are entertained from the coming of so many scientists to the Pacific Coast.

R. H. Freund demonstrated spirilla, showing flagella. He exhibited a slide of rare excellence.

PERSONAL.

Walter F. Webb formerly with *The Oologist* has started a monthly magazine called *The Museum.* The first number contains a finely illustrated article upon the Atlantic Coast Starfishes.

H. C. Wells is an authority on life slides. We hope to have an article from him on this subject.

D. Bryce Scott is interested in the Polycistina of which there are but few students in America. He sometimes has earth for distribution.

Dr. Geo. S. Liggett has three little crosses cut in his stage and uses them as a "finder" by putting a speck of ink on the glass, right over the crosses.

Hans Wilder is a successful druggist who knows what to do with a microscope.

Albert S. Barker has become assistant editor of the *Bulletin.*

Lewis Woolman has samples of a small insect which in countless numbers rode from New Jersey into Philadelphia on a locomotive. The wings are prettily veined and the eyes multiple (family Leptoceridæ).

F. W. Richards is an enthusiastic member of the Montreal Microscopical Society.

Dr. P. W. Shimer has invented an apparatus for filtering microscopical objects out of water.

B. H. Gledhill has a polarizing arrangement giving a great number of color combinations. Will he please send us a full description?

Wm. R. Huggard of Davos Platz, Switzerland, has contributed a valuable article on Consumption to the *Universal Medical Journal.*

Dr. Chas. E. Page thinks that typhoid can be best treated by fasting and using the brand bath.

George Otis Mitchell is the secretary of the San Francisco Society who used to send us reports of its meetings.

A. N. Edwards has decided not to publish the book on Bacilariaceæ at present owing to the hard times and the consequent economy of subscribers.

Dr. Albert Schneider is one of the rising biologists of Minnesota. He is studying Symbiosis.

Dr. W. W. Alleger will be pleased to know that his paper on formalin is being quoted quite extensively.

Alfred Allen prefixes a preface to his index for 1894. This is a new departure in magazine work and quite a good one.

Dr. A. C. Stokes has an article on the structure of insect tracheæ in the current number of Alfred Allen's journal. Better make him one of the associate editors, brother Allen.

NEW PUBLICATIONS.

Travaux d'Electrotherapie Gynecologique. Par Le Dr. G. Apostoli. Paris 1894, 8° pp. 720.

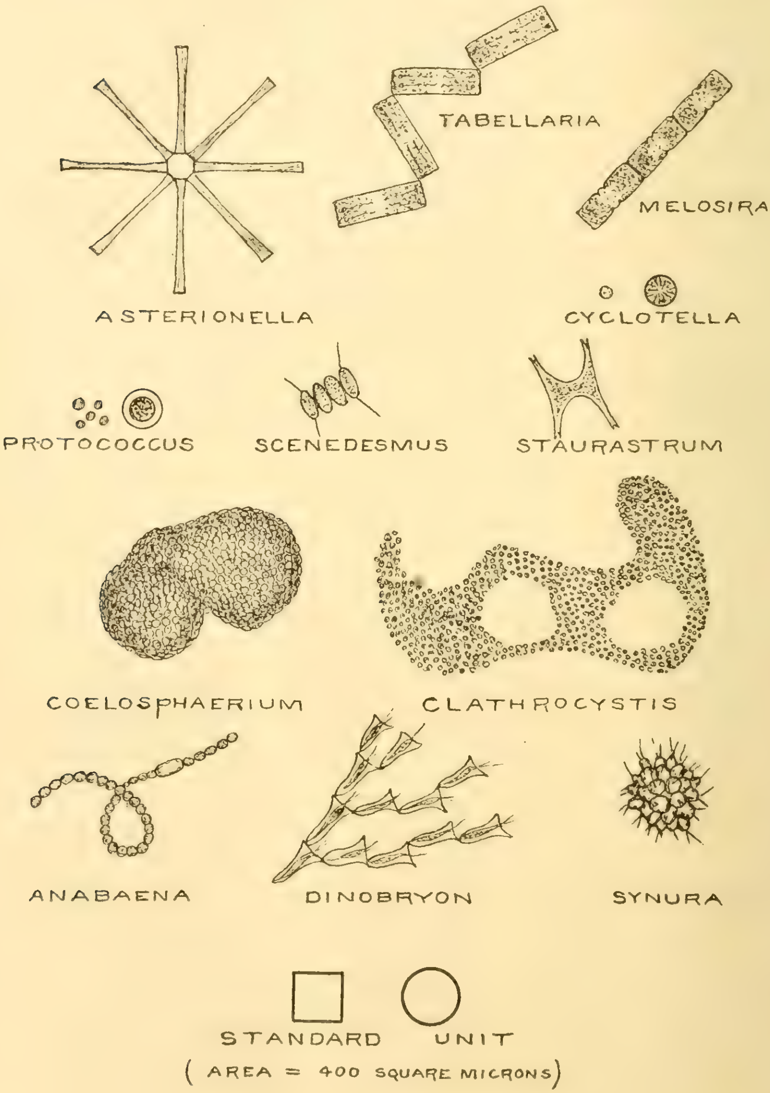
We have received Vol. 1, Nos. 1 and 2 of this semi-annual periodical and find it to contain an immense amount of matter collected from the publications of the past 10 years upon this specialty. The works of medical congresses and of societies have been largely drawn upon. England, Belgium, America, Russia, Italy, Germany, Austria, Denmark, Poland, Hungary and Canada are represented. It will be of great value to Gynecologists.

Report on Typhoid Fever in the District of Columbia submitted by the Medical Society of the D. C. to Congress, June 14, 1894.

This document sets forth an alarming prevalence of typhoid for several years past, attributes them to infected milk and water. The Potomac water is not badly infected, but it is the water of public wells which is accused. They advise closing them all regardless of protests by the masses whose only dependence in hot weather for a cool drink is upon these wells. Water of the river gets warm and contains mud. Altogether, the outlook is serious.

RELATIVE SIZES

OF SOME COMMON MICRO-ORGANISMS.



THE AMERICAN
MONTHLY
MICROSCOPICAL JOURNAL.

VOL. XV. DECEMBER, 1894. No. 12.

Haematoblasts and Blood Platelets.

By DR. M. L. HOLBROOK,
NEW YORK.

[Read before the American Society of Microscopists in Brooklyn, N. Y., August 1894.

Although a large number of studies and publications have been made during the last 20 years, on the morphological elements of the blood, we have not yet reached a thorough understanding of the significance of certain form-elements known as the so-called "third corpuscles." In my studies of the human blood, extending over several years, I have reached some conclusions which I wish to lay before this society. The reasons why great discrepancy of opinion on certain points prevails among observers are two fold. First the structure of the red blood corpuscles is not agreed upon by the majority of microscopists; second, under the term— "third element" both hæmatoblasts and blood platelets have been included by most authors who have written on the subject. I have in the last two years laid before you observations which I think go far to prove the correctness of Elsberg's assertions made in 1879, that the structure of the red blood corpuscle is reticular, the same as that of the colorless blood corpuscles, the difference being, that in the former, the meshes of the recticulum are filled with a chemically complicated structure, generally termed hæmoglobin, whereas, in the latter, the meshes hold a colorless nitrogenous liquid, the "parablast" of French microscopists. It seems to me that no understanding of

the so-called third element is possible, unless we admit the reticular structure of the red blood corpuscles. As to the second point, it will be the topic of the present paper. I wish to state beforehand that hæmatoblasts and blood platelets are not identical formations, but of widely different origin and significance. As to the method of investigation, I wish to emphasize, as I have done in previous years, that reliable observation is possible, only when we take blood directly from the skin and with as little delay as possible and transfer it immediately to a moist chamber under the microscope. I need not again describe what I have done in previous years—how to prepare specimens, but will add that I place emphasis upon the size of the droplet of blood to be taken as well as the rapidity of transferring it.

The size of the droplet should be at least as large as a medium-sized pin's head. Such a droplet will spread uniformly under a three-quarter inch cover glass, whose edges have beforehand been oiled. Smaller drops do not spread so evenly, and the thin places are apt to dry up under the influence of the air inclosed under the cover. No time should be wasted in squeezing out a droplet too small from too slight a wound with the needle. A few seconds delay will suffice to cause crenation of the majority of the red blood corpuscles, even though in summer heat; whereas, the contour will be almost uniformly smooth, if there is no delay in removing the blood into the moist chamber of the slide. I object to the addition of any reagent such as a solution of osmic acid, Pachini's preserving liquid, or even a solution of table salt; they all quickly alter the appearance of the blood and lead to misconceptions. Nothing but the serum of the blood itself should be allowed to bathe the blood corpuscles. In it alone as I have already shown in a previous paper they may be preserved for several days.

I.—HÆMATOBLASTS.

Hæmatoblasts are shining or lustrous homegeneous globules or disks of a marked yellow color, measuring 1.5 and 3.5 micromillimeters in diameter up to nearly the size of fully developed red blood corpuscles. They are juvenile forms of the red blood corpuscles present in large numbers in the blood of the newly born and in small numbers in the blood of healthy adults. I have seen them in small numbers in the blood of a healthy man 63 years of age. They are invariably augmented whenever there has occurred a hemorrhage of some extent, as also after frequent slight hemorrhages. We see them with great regularity in urine holding red blood corpuscles to a varying number due to hemorrhages induced by tumors of the bladder, especially benign papiloma, or hemorrhages from the pelvis of the kidneys due to so-called renal calculi. Their number varies according to the severity and frequent repetition of the hemorrhages. I have seen them in numbers exceeding the number of the red blood corpuscles. They have nothing to do with wasting diseases such as tuberculosis, cancer, syphilis, etc. Neither are they augmented in numbers in chlorosis.

This I think goes far to prove that hæmatoblasts are juvenile forms of red blood corpuscles and their most characteristic feature is their yellowish color due to the presence of hæmoglobin even in the earliest recognizable stages when they do not exceed the size of a granule. I have stated before that hæmatoblasts are homogeneous and apparently structureless. This is true only of the smallest forms.

As soon as they have reached a diameter of about 3 micromillimetres with a power of the microscope of 1,000 to 1,200 diameters, we recognize in them an extremely narrow and faint reticular structure after their exposure to a solution of 1-5 of 1 per cent of chromic acid, or a 40 to 50 per cent of saturated solution of bichromate of potash.

The latter brings forth distinctly the reticular structure of fully developed red blood corpuscles. The smallest hæmatoblasts under the influence of these solutions are either unchanged or exhibit a small central or excentric vacuole whereby the hæmatoblast is rendered ring shaped.

Hæmatoblasts up to half the diameter of a red blood corpuscle resist, at least for some time, a solution of glacial acetic acid with equal parts of water. They retain both a yellow tint and a smooth contour, whereas fully developed red blood corpuscles by this reagent are either rendered flat, much enlarged, corrugated into many shapes, or broken up as will be described later.

The smallest hæmatoblasts always retain a smooth surface, even though the red blood corpuscles in the same specimen have become crenated all over. When the hæmatoblasts have reached about half the size of fully developed red blood corpuscles they assume a finely crenated surface, the crenations appearing like needle shaped projections from their margins, and not blunt, or knob-like, as are those of fully developed red blood corpuscles.

As to their origin I shall not make positive statements for the present. I am sure however, that the old opinion as to the origin of red blood corpuscles from colorless corpuscles is erroneous. Haematoblasts are formed in the tissues of the embryo, or the fœtus whenever one type of connective tissue changes into another, for instance, cartilage into bone. Haematoblasts are formed whenever in the process of development of the tissues new blood vessels arise. They are formed in the adult in all lymph structures, mainly in the lymph ganglia and in the spleen throughout life. The older a person grows the less of lymphatic structure is present in the organism, hence haematoblasts are found only in small numbers in the blood of very aged people. Final statements as to the origin of haematoblasts I leave until I have fin-

ished some studies of the *area vasculosi* of the embryo of the chicken.

Hæmatoblasts are granules or isolated lumps of living matter not yet of the size of red blood corpuscles. They are early forms of red blood corpuscles and develope into such. They are homogenous if small, the size of granules say 1.5 *micro millimeters and the more distinctly reticular in structure the nearer they reach the size of red blood corpuscles. They are from the very beginning saturated with haemoglobin.*

Most writers quote Hayem as the discoverer of these bodies which is erroneous, for neither the name haematoblast nor an accurate description of them can be credited to him. Foster's Medical Dictionary is, as far as I know, the only work mentioning C. Heitzmann as the originator of the name. (See article Haematoblast in this work.) This author defines a hæmatoblast correctly when he says they are miniature red blood corpuscles. This term is applied also to small, colorless, circular or ovoid bodies, 1-2 to 1-6 the size of red blood corpuscles circulating in the blood of mammals. It is, according to Foster, probably identical with the Hæmatoblast of Hayem and the blood plate of Osler. This is an error, the same one Hayem himself made when describing the hæmetoblasts in 1877.

Heitzmann ("Studien am Knochen und Knorpel, Jahrbucher, 1872,) described the changes occuring in the hyaline cartilage preceeding the formation of bone tissue. At the border of the hyaline cartilage may be found isolated lumps of living matter with high refraction, of a yellowish color, evidently arising from previous cartilage corpuscles. He says " as the shining solid corpuscles exhibited stages of development advanced to the formation of nearly perfect red blood corpuscles, I considered them to be juvenile forms of the latter and propose for their designation the term *hæmatoblast.* I con-

cluded that a part of the cartilage corpuscle, or a portion of the body of such a corpuscle, had become transformed into hæmatoblasts." His statements are illustrated and have been corroborated by Kassowits and others. In 1873 another paper appeared by this author in which the new formation of blood vessels is described in the process of transformation of cartilaginous into bone tissue, likewise with numerous illustrations in which the hæmatoblasts are seen lying in vacuoles of elongated formations of living matter, the future blood vessels. He claims that both blood corpuscles and blood vessels originate simultanously, the former from isolated lumps of living matter which he termed hæmatoblasts; the latter from elongated tracts of living matter hollowed out by vacuolation and the accumulation of a liquid, in which the hæmatoblasts are suspended. From this it is evident that—Heitzmann was the discoverer of the hæmatoblasts; the first to correctly describe them, and the first to give them a name.

In 1877, G. Hayem presented to the Academy of Sciences of Paris, in brief succession four short papers published in the Comptes Rendus, Vol. 84, which relate to the red blood corpuscles. In the first paper he spoke of the anatomical features of the blood of the new born. He states that the sizes of the corpuscles are more varied than in the blood of the adult.

In the second paper he discusses the nature and significance of the small blood corpuscles. He contradicts a prevalent opinion that these corpuscles indicate a retrograde change, or atrophy; on the contrary, he concluded from a number of facts which he observed in convalescent persons after acute wasting diseases, in anæmia, in menstruation, etc., that these small elements which he terms dwarfed corpuscles are young red blood corpuscles incompletely developed.

In a third paper upon the development of the red

blood corpuscles of the blood of oviparous vertebrates, he says; "the blood of oviparous vertebrates constantly holds colorless cells differing essentially from white blood corpuscles. These elements in progressive development become perfect red blood corpuscles and for this reason he proposes to call them by the name of Hæmatoblasts."

In the fourth paper upon the development of the red corpuscles in the blood of higher animals, viviparous vertebrates, he states that the hæmatoblasts in the blood of men and viviparous vertebrates are very small, delicate, slightly refracting elements of a contour almost invisible, with a diameter of 1-5 to 3 micromillimeters. Immediately after the removal of the blood from the vessels the hæmatoblasts become thorny and are apt to form groops or clusters, which latter feature is less pronounced in the blood of man than in certain animals. He thinks that they play an important part in the formation of fibrin, though this is doubtful. In his conclusions, he states that the red blood corpuscles arise from more or less regular small elements which are colorless, delicate and quickly changed or modified as soon as removed from the blood vessels of living creatures.

From the description of Hayem I think it plainly follows that he designated as hæmatoblasts two form elements which I hope to show are entirely distinct from each other.

II.—BLOOD PLATELETS.

The first one who saw in the fresh blood granules, granular masses or plaques was Max Schultze. [Archive fur Mikroscopische Anatomie. Vol. 1, p. 38, 1861.] Today instead of the word plaque we adopt the name platelet, it being the diminutive of plate, identical with the French plaque.

Blood platelets are colorless lumps of living matter

varying in size from just visible granules up to the size of about ½ the diameter of a red blood corpuscle.

The smallest granules are, as seen under immersion lenses, apparently homogeneous or structureless, the larger the granules are the more we are able to discern in them an extremely delicate faintly pronounced reticular structure. All these formations I have stated are colorless, exhibiting when small a peculiar greenish shining appearance characteristic of all granules of living matter.

They invariably lack the yellowish color seen in the Hæmatoblasts. Small granules are nearly uniform in their diameter and are often seen slightly angular, *i. e.*, provided with from two to four conical projections. The larger the granules swell owing to their taking in of liquid from the surrounding plasma of the blood, the more they assume a grayish color, a discoid shape, smooth contours and a faint reticular structure.

The easiest way to bring the platelets to view is to treat the blood corpuscles with glacial acetic acid, diluted with equal parts of water. For this purpose a droplet of the solution of this acid is placed upon the skin, preferably the palmer surface of the thumb of the left hand. The skin is then pricked directly through this droplet. Both the blood secured and acetic acid are directly transferred to the slide and quickly covered with a thin covering, glass always of course oiled on its edges. In my own blood platelets are extremely scanty. As soon, however, as acetic acid is mixed with it hundreds of them appear, of all sizes from the minutest granules, up to half the size of a red blood corpuscle, the smallest being slightly angular, glossy and of a greenish refraction ; the larger ones pale gray, smooth, discoid and faintly reticulated. The red blood corpuscles, under the influence of this preparation of acetic acid in a few minutes have become enlarged to nearly three times their original size,

much flattened and deprived of their coloring matter. The experiment I have here described was first made by Norris, of England, who thought he had discovered a new element in the blood, and proposed the awkward name "Invisible corpuscles of the blood." He evidently did not take into account the fact that the platelets were artificial products caused by the breaking up of the blood corpuscles. Norris's experiment is valuable, however, inasmuch as it enables us to produce them in large numbers in a short time.

The question now arizes, what are platelets? I may state that without doubt they are products of the red blood corpuscles caused by their breaking up into fragments of varying sizes. My reasons for this belief are the following:

1. In specimens of fresh blood transferred at once to the microscope I have seen, when the red blood corpuscles were distinctly crenated, knobs hanging upon the corpuscles by means of a slender pedicle. These knobs, after the pedicles break, float freely in the surrounding serum, exhibiting a slightly angular contour and all the features of blood platelets.

2. If the blood of persons with an impaired constitution is prepared and kept for a few days being examined daily we shall in a short time, beginning perhaps on the second or third day, notice clusters of platelets which judging from their grouping, must have arisen from the breaking up of red blood corpuscles.

3. If we treat fresh blood with glacial acetic acid diluted with equal parts of water, the red blood corpuscles are immediately transformed into large flat and pale plates as already described, with blood platelets varying in size from the minutest granules up to those of ordinary size. At the same time, in the surrounding blood plasma numerous platelets have appeared.

4. The observations that platelets arise from red blood corpuscles had already been made. L. Elsberg after the treatment of red blood corpuscles with a 40 to 50 per cent solution of bichromate of patash. (Annals of the New York Academy of Sciences Vol. 1875.)

He says, "An indentation at the periphery of the red blood corpuscles is due to locally limited contraction of the net work in the interior of the corpuscles. Contraction of the living matter in one part of the periphery will bring about a protusion of the flap at another, the flap being bounded by the layer of the corpuscle. Segmental contraction of the net work will produce a rupture of the outer layer of the corpuscle, with projection of a pediolated granule or knob, formerly a part of the interior network ; contraction will be followed by the rupture of the pedicle and the production of either so called detritus, or small granules, or when the protuded knob is larger, or has become swelled, of a pale grayish disk.

L. Elberg adds to this remarkable observation for which he never has been given credit, the following remark. "The peculiar corpuscles believed to be characteristic of syphilis by Lostorfer, and proved by Stricker to be present in the blood of individuals broken down by that and various other diseases are nothing but such disks, *i. e.*, portions of the colorless blood corpuscles protruded from the interior, detached and more or less swelled. As persons in low states of health have a relatively small amount of living matter in the same bulk, or in other words, only a delicate net work within a protoplasmic body, or plastid, the so called cell, such net work suspended in a relatively large amount of fluid can much more easily contract and bring about a rupture of the outer layer, than in the case of healthy persons within whose plastids there is relatively less room for contraction to take place."

All the named observations explain why a platelet has

no color and holds no hæmoglobin. If we recall the fact that a red blood corpuscle is made up of a reticulum of colorless matter, in whose meshes is suspended more or less hæmoglobin, we readily understand that the platelets being exclusively formations of living matter lack hæmoglobin altogether.

While I am positive that the platelets are products of the red blood corpuscles, I cannot deny that they also arise from colorless blood corpuscles. I have made several observations which indicate that they also originate from granules or rather points of intersection of the reticulum of living matter of colorless blood corpuscles. Platelets under all circumstances are identical with granules of living matter.

As to the significance of the blood platelets I do not claim that they are of pathological significance. This much, however, is certain, that in the blood of perfectly healthy persons they are either very few or entirely absent. On the contrary, the more a person's health is impaired by any chronic disease the more certain we are to find a large number of them in his blood, when taken from the vessels a few seconds before microscopical examination. In persons suffering from Syphilis, Cancer, Tuberculosis, or even from Neurasthenia we find them in considerable numbers. Lostorfer, (Medicinische Jahrbucher, 1871,) was as he admitted mistaken in bringing the platelets into relation with Syphilis. R. L. Watkins, (The Medical Brief Vol. 21, 1893,) is, I hold, also mistaken in bringing the platelets into exclusive relation with tuberculosis. No doubt these bodies are always present in large numbers in the blood of persons with other chronic wasting diseases.

Wm. Osler in his Cartwright lectures, 1886, claims that the so called third corpuscle is identical with the Hæmatoblast, as most other observers do. He quotes Hayem as the discoverer of the hæmatoblast as well as of the

blood platelets. I have drawn attention to the fact in the first half of my paper that Hayem is not the discoverer of the haematoblasts, and now I will also add that Hayem was in error in confounding the haematoblasts with blood platelets.

Osler's views are marred by the acceptance of Hayem's error. There can be no doubt that Lostorfer in 1871 first saw the platelets although he misinterpreted their significance. We may call Max Schultze, the discoverer of the granular masses in the blood, though this excellent observer had in 1860 no idea of the life and structure of the red blood corpuscles which he considered as merely chemical constituents of the blood.

Julio Bizzozero "(Archiv. Ital. de Biol., 1882)" states that the platelets which he terms plaques serve as centers of coagulution of the fibrin and play an important role in the formation of the white or fibrous clot of the thrombus obliterating the calibre of the injured blood vessel, forming masses that constitute the chief element in the white or fibrinous thrombus. Recent studies seem to be opposed to this view.

Many observations have proved that both Haematoblasts and blood platelets circulate in the vascular system of living animals.

If we consider the fact that healthy persons have, in their red blood corpuscles a compact reticulum of living matter, whose meshes are filled with haemoglobin, while debilitated persons, on the contrary have a delicate reticulum in their red blood corpuscles and comparatively little haemaglobin which is probably also of less consistance than in vigorous persons, we are at once in the position to understand the origin and the significance of the platelets. The living matter during the life of the organism is at no time in perfect rest. Perfect rest means death. Contractions of the living matter in the red blood corpuscles of a healthy person will have but

little effect and will rarely cause the protrusion and detachment of platelets. The more delicate the reticulum the more effective will be the result of its contractions, the more so if haemoglobin is scarce and of a more liquid consistency. Contraction of the living matter in this instance will easily yield knob-like protrusions over the periphery of the red blood corpuscles which knobs being detached will appear in the shape of platelets floating in the serum of the blood.

In my judgement the presence of platelets in the blood in large numbers is not a pathological feature as such, but merely an indication of a pathological condition of the system. Every chronic disease that causes a wasting of the living matter and a decrease in the amount of haemoglobin will cause the appearance of platelets in considerable numbers within the vascular system.

A Note on the Use of Anise Oil in Histological Methods with Special Reference to its Value in Cutting Serial Sections on the Freezing Microtome.

BY VERANUS A. MOORE, M. D.,
WASHINGTON, D. C.

In 1892 Kuhne(*) called attention to the value of anise oil as an embedding medium for animal tissues to be cut on a freezing microtome. The process recommended by him was to fix the tissue in alcohol, after which small pieces not to exceed one or two millimeters in thickness are placed in absolute alcohol for a few hours, after which they are immersed in anise oil for from 12 to 24 hours. At the expiration of that time they are ready for sectioning. The piece to be cut is prop-

*Centrablatt f. Bakteriologie u. Parasitenkunde, XIII.

erly arranged on the table of the freezing microtome, covered with a few drops of anise oil, and then frozen by means of an ether spray or any of the other freezing appliances.

The advantages claimed for this method are ; (1) the sections can be cut by the freezing process from alcoholic specimens ; (2)the edge of the section knife is not injured by the congealed oil, as invariably follows the section of tissues frozen in an aqueous solution; and (3) the tissue is frozen much more quickly and when congealed it remains so much longer than when frozen in the ordinarily used media which enables a larger number of sections to be cut at a single freezing. The sections are removed to alcohol, hydrated, stained, dehydrated and mounted as if cut by the paraffin method. The necessity of a thoroughly practical process by which tissues can be cut from alcoholic specimens and prepared for microscopic examination in a short time is keenly felt especially in pathological investigations. The method suggested by Kuhne, therefore, filled a much needed want in this particular. The time saved, however, is not so great as at first imagined, but the elimination of the paraffin infiltrating process is sometimes especially desirable.

More recently Coats (*) has applied anise oil as a medium for holding blocks of tissue to the table of the freezing microtome without previously infiltrating them with the oil. He does not find the anise oil satisfactory as an infiltrating substance, but highly recommends it as a medium in which to freeze tissues. He proposes a modification of Kuhne's application of this substance which materially shortens the time necessary to section and mount tissues for examination. His method is as follow :

The tissue or organ from which sections are to be made is cut into small blocks 2–4mm. in thickness and

*The Journal of Pathology and Bacteriology, II. (1894), p. 492.

correspondingly small on the superficial surface. These are placed in a test tube containing absolute alcohol. The blocks are so placed in the tubes that they lie perfectly flat upon a layer of absorbent cotton in the bottom of the tube. The tube is placed in a water bath and raised to a temperature of about 40° C. If the tissue is large the alcohol should be changed after a short interval. In the course of about one-half hour the tissue will be sufficiently hardened to be proceeded with further.

The block of tissue to be cut is removed from the alcohol, dried with a piece of blotting paper and placed on the table of the microtome, covered with a few drops of anise oil and frozen as recommended by Kuhne. The sections are subsequently stained and mounted. By this process fresh tissues may be sectioned within an hour after removing them from the body.

After the appearance of Kuhne's article, I used his method with very satisfactory results. Contrary to Coats' experience I found the infiltrated tissue to cut better than when it was simply hardened and to stain quite as satisfactorily. These points of difference, however, are unimportant as either of the processes afford a means of rapid preparation of alcoholic tissues. The use of anise oil as a medium in which to freeze the tissue is, on account of the features already mentioned, to be highly recommended.

In applying Kuhne's method the idea was suggested that where for any reason several sections were to be mounted from the same block of tissue, time could be saved by staining the tissue *en masse*. The fact that anise oil and Canada basalm are nuisible renders it possible to mount the sections directly from the section knife when the tissue has been thoroughly infiltrated with the oil. This eliminates the after treatement of the sections and materially shortens the ordinary process.

The above facts render the use of anise oil of superior

merit when serial sections are to be cut. The tissue can be hardened, stained *en masse* as by ordinary methods. The infiltration with anise oil can be accomplished without injury to the stain ordinarily used, and when cut the section canbe transferred directly to the slide, and mounted in balsam at once. The ease with which tissues infiltrated with anise oil can be frozen and the length of time the tissue remains congealed enables one to continue cutting as long as desirable. A constant pressure apparatus, such as a carbonic acid gas cylinder, is. of course preferable for freezing although an ordinary ether spray apparatus can be used. As the congealed anise oil does not injure the edge of the knife there is little or no danger in using the knife employed in cutting paraffin sections.

A scheme of procedure for putting up sereal sections by this method is as follows:

Fix the tissue in alcohol.
Stain *en masse*. (ordinary method.)
Dehydrate. (Absolute alcohol.)
Infiltrate with anise oil.
Section on freezing microtome.
Mount in balsam.

The efficieucy of this process for delicate histological work is questioned but for many of the purposes for which it is desirable to make sections it affords a very satisfactory method for obviating the paraffin process. Its value lies in the rapidity with which sections can be made from alcoholic specimens where the object to be attained can be accomplished with low power objectives. As suggested by Kuhne and Coats the use of anise oil is largely restricted to certain kinds of pathological work where it is occasionally very important to have a method whereby the results of a somewhat crude microscopic examination cau be obtained in a very short time.

A Standard Unit of Size for Micro-Organisms.

BY GEORGE C. WHIPPLE,

BIOLOGIST, BOSTON WATER WORKS.

WITH FRONTISPIECE.

The Sedgwick-Rafter* method of microscopically examining water is now in quite general use. Experience has proved that it is a most satisfactory and valuable means of studying the micro-organisms found in water. But in spite of the excellent precision† of the methods, the results of the examinations as they are generally stated are sometimes quite misleading; that is, they do not always correctly represent the amount of animal and plant life present in the water. It has usually been the custom of biologists to record the number of organisms present in a cubic centimeter of the water examined, without regard to their character or size; single cells, filaments, colonies, irregular masses have been counted as units and the same value assigned to each.

This was the method first used in the Biological Laboratory of the Boston Water Works. The results were, in some respects, unsatisfactory. When one compared the number of organisms found at the different seasons of the year it was found that the largest numbers did not occur at the times when the water, judged by its appearance, taste and odor, was manifestly the poorest.

Furthermore, the microscopical results did not always compare well with the chemical analyses. This was found to be largely due to the different sizes of the organisms. Coelosphaerium, for instance, was found to contain more than a thousand times as much organic

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*W. T. SEDGWICK.—Report of the Biological Work of the Lawrence Experiment station. Special Report of the Massachusetts State Board of Health on the Purification of Sewage and water 1890.

GEO. W. RAFTER.—The Microscopical Examination of Potable Water. Van Nostrand Science Series, No. 103.

†GARY N. CALKINS.—The Microscopical Examination of Water, 23rd Annual Report of the Massachusetts State Board of Health, 1891.

matter as a small Cyclotella; and, according to the method used, these were given equal weight in the report.

These facts led us to seek some unit in terms of which we could express, as nearly as possible the actual quantity of animal and plant matter present. After a long investigation it was decided to adopt the standard already in use for the estimation of the amorphous matter, namely, a square twenty microns on a side, having an area of four hundred square microns. This, when modified in case of organisms known to be either very thick or very thin, gave us practically a unit of volume. This modified unit of area probably approaches as near to an exact volumetric unit as it will be found practicable to use.

The area of four hundred square microns was selected as the standard unit because it had already been used in estimating the amorphous matter, and our observers had therefore become accustomed to it; because it was about the size of several of our most common organisms; and because it was a unit whose size could be easily carried in the mind. This unit can be used with but little extra labor. Many organisms are very constant in size; these may be counted, as heretofore, and reduced to the new standard by multiplying by a previously determined factor. Filamentous forms which are of a constant width may be measured in length and then reduced to standard units. In the case of irregular masses, and of organisms and colonies which vary in size, a special estimate must be made for each. After a little practice this can be done very quickly and accurately. It will be found of great advantage to have the ocular micrometer divided as follows :—the square, which should cover one square millimeter on the stage of the microscope, is first divided into four equal squares and each of these quarters sub-divided into twenty-five smaller squares, each of which is equivalent to twenty-five standard units.

The eye will readily divide the side of a small square into fifths, and this division will be the side of the standard unit square. Thus the size of the unit may be kept constantly before the eye of the observer.

Table which will be found useful in estimating the number of standard units in filamentous algæ.

	Diameter of Filament in microns.	Number of Standard Units for* each hundred microns of length.
No. 1	0–5	0.8
2	5–10	2.0
3	10–20	4.0
4	20–30	6.0
5	30–40	8.5
6	40–50	11.0
7	50–60	13.5
8	60–70	16.0
9	70–80	18.0
10	80–90	20 0

List showing the ordinary value, in standard units, of some of the organisms found in the Boston Water Supply.

DIATOMACEÆ.

Asterionella.	0.4	Navicula.	0.2 to 1.0
Cyclotella.	0.1 to 1.0	Stephanodiscus.	1.0
Diatoma.	0.3	Synedra.	0.2 to 2.5
Fragilaria.	0.8	Tabellaria.	0.9
Melosira.	0.5		

DESMIDIEÆ.

Closterium.	0.8 to 6.0	Xanthidium.	3.0
Staurastrum.	1.8	Micrasterias.	6.0
Cosmarium.	3.8		

CHLOROPHYCEÆ.

Coelastrum.	4.0	Protococcus.	1.2
Gonium.	2.0	Raphidium.	0.8
Pandorina.	7.0	Scenedesmus.	0.3
Pediastrum.	5.0		

CYANOPHYCEÆ.

Anabaena.	2.0 to 5.0	Coelosphaerium.	5.0 to 50.0
Chroococcus	1.3	Microcystis.	2.0 to 10.0
Clathrocystis.	10.0 to 100.0		

FUNGI.

Crenothrix.	2.0	Beggiatoa.	2.0

*The smallest squares of the micrometer are one hundred microns on a side.

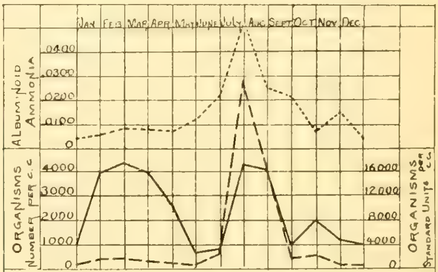
RHIZOPODA.

Amoeba.	4.0	Difflugia.	7.0
Actinophrys.	3.0		

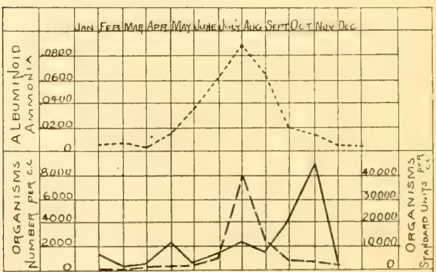
INFUSORIA.

Codonella.	6.0	Peridinium.	3.5
Cryptomonas.	1.0	Synura.	2.0 to 10.0
Dinobryon.	0.5	Tintinnidium.	7.5
Mallomonas.	1.0	Trachoelmonas.	1.0

In order to show the difference between the Unit Method and the old method of counting, the accompany-



LUDLOW RESERVOIR
SPRINGFIELD, MASS. 1890.



HAYNES RESERVOIR
LEOMINSTER, MASS. 1890.

ing plate was prepared. The profiles were drawn from the analyses of the Massachusetts State Board of Health. Ludlow Reservoir, Springfield, and Haynes Reservoir, Leominster, were selected because they showed great seasonal variations in the number and kind of organisms. The full line shows the number of organisms per c. c. as given in the report. The broken line represents the

same organisms expressed in terms of the standard unit. The dotted line represents the "suspended albuminoid ammonia" in parts per 100,000. It will be noticed that there is a general parallelism between the profile of the albuminoid ammonia and that of the organisms expressed in terms of the standard unit. This parallelism is what ought to be found, because the suspended albuminoid ammonia is supposed to represent that part of the nitrogenous matter present as organisms or fragments of living of dead matter, held in suspension in the water. It will also be noticed that the curve of organisms plotted according to the actual numbers, does not correspond well with the chemical analyses.

In conclusion it may be stated that this volumetric method has been used in the Biological Laboratory of the Boston Water Works since Jan. 1, 1893*, and has given excellent satisfaction. It is also in use in the Biological Laboratory of the Lynn Water Works, and several biologists in other laboratories have signified their intention of adopting it.

Classification of the Radiolaria; Key to the Species of Barbadoes.

BY REV. FRED'K B. CARTER,

MONTCLAIR, N. J.

This Key has been constructed from the descriptions of the species of Barbadoes which are scattered throughout the two huge volumes of text of Haeckel's great work on the Radiolaria.

1. CENOSPHÆRA.

Pores circular, with hexagonal frames, 14–16 on the quadrant.........melecta.
Pores irregular, roundish, 30–40 on the quadrant..........................gigantea.

2. ETHMOSPHÆRA.

Pores prolonged into conical tubuli, 16–18 on the quadrant......polysiphonia.

*18th Annual Report, Boston Water Board, 1893, page 72.

3. CARPOSPHÆRA.

Pores hexagonal, 5–6 on the quadrant.......................................infundibulum.
Pores circular, 6-8 on the quadrant.......................................entactinia.
Pores circular, 8–12 on the quadrant....................................... modesta.
Pores irregular, roundish, 2–4 times as broad as the bars....................nobilis.

4. THECOSPHÆRA.

Pores circular, hexagonally framed ; shells connected by 12 radial beams ....favosa.
Pores circular ; shells connected by 4 radial beams........................... medusa.

5. RHODOSPHÆRA.

Pores irregular, roundish ; diameter of shell 0.4..............................palliata.

6. CROMYOSPHÆRA.

Pores circular ; surface thorny..cepa.
Pores irregular, roundish ; surface smooth...............................scorodonium.

7. CARYOSPHÆRA.

Pores circular ; six concentric spheres.......................................hexalepas.

8. XIPHOSPHÆRA.

Pores circular, 15–20 on the half equator ; bristle like spines...............juno.
Pores circular, 28–32 on the half equator ; conical thorns....................gigantea.

9. XIPHOSTYLUS.

Pores circular, 8–10 on the half equator.......................................anhinga.

10. SATURNALIS.

Pores circular, 8–10 on the half equator ; ring without spines or thorns.cyclus.
Pores circular, 12–16 on the half equator ; ring with spines or thorns..trochoides.

11. STYLOSPHÆRA.

Pores circular, 10–15 on the half equator ; polar spines prismatic......hispida.
Pores circular, 8–10 on the half equator ; polar spines conical. .....liostylus.

12. SPHÆROSTYLUS.

Pores circular, 10–12 on the half-equator.....................................liostylus.
Pores circular, 8–10 on the half-equator ; polar spines S like.........flexuosus.

13. STAUROSPHÆRA.

Pores circular, 20–22 on the quadrant ; surface smooth.......................petri.
Pores circular, 8–10 on the quadrant ; surface smooth.......................simonis.
Pores irregular, roundish, 4–6 on the quadrant; surface smooth.apostolorum.
Pores irregular, roundish, polyonally framed, 5–7 on the quadrant ; surface spiny.. thaddæi.

14. STAUROLONCHE.

Pores circular, 4–5 on the quadrant; surface smooth........................aperta.
Pores circular, 20–22 on the quadrant; surface spiny.................feuerbachii.

15. STAUROLONCHIDIUM.

Pores circular, 3–4 on the quadrant.. perspicuum.

16. STAURACONTIUM.

Pores circular, 16–18 on the quadrant... ....sparganium.

17. HEXALONCHE.

Pores circular, hexagonally framed, 6--8 on the radius.......................favosa.

18. HEXACONTIUM.

Pores irregular, roundish, 5--7 on the radius......... ....setosum.

19. ACANTHOSPHÆRA.

Pores circular, twice as broad as the bars; 30--40 radial spines .....compacta.
Pores irregular, roundish, 7--9 on the radius, 10--20 radial spines ..acanthica.

20. HALIOMMA.

Pores circular, 8--10 on the radius; commonly spines at all the nodal points ....horridum.
Pores circular, 6--8 on the radius; spines not at all the nodal points..oculatum.
Pores irregular, roundish, about 20 on the radius; commonly spines at all the nodal points..... ....rhodococcus.

21. ACTINOMMA.

Pores irregular, roundish; diameter of shell 0--4............................ giganteum.

22. ECHINOMMA.

Pores circular, 8--10 on the radius..aculeatum.

23. CENELLIPSIS.

Pores circular, 8--9 on the half-equator......................................ehrenbergii.

24. ELLIPSIDIUM.

Meshes hexagonal, 18--20 on the half-equator.........................pandanidium.

25. ELLIPSOXIPHUS.

Meshes hexagonal, 8--9 on the half equator......... ....flosculus.

26. ELLIPSOSTYLUS.

Meshes irregular, roundish, 8–10 on the half-equator.....................hirundo.

27. LITHOMESPILUS.

Pores irregular, roundish, 10–12 on the half-equator........................flammeus.

28. DRUPPULA.

Pores hexagonal, 12--15 on the half-equator..drupa.

29. PRUNOCARPUS.

Pores circular, very small, 40–50 on the half-equator. .... spargianum.

30. LITHATRACTUS.

Meshes circular, 10–12 on the half-equator; surface thorny..............carduelis.
Meshes irregular, roundish, 6–8 on the half equator, surface smooth. . lobatus.

31. DRUPPATRACTUS.

Pores circular; surface thorny or papillose......................................coronatus.
Pores irregular, roundish; surface smooth ....lævis.

32. STYLATRACTUS.

Pores circular, 10–12 on the half equator ....carduus.

33. XIPHATRACTUS.

Pores circular, 7–8 on the half-equator..spinulosus.
Pores circular, 9–10 on the half equator; polar spines furrowed......sulcatus.
Pores irregular, roundish, 9–10 on the half-equator....................... radiosus

Three of the above genera were omitted from the key to the genera, namely, *Caryosphæra*, *Ellipsostylus* and *Druppula*.

Caryosphæra should be inserted (on p. 226 of the Journal, August 1893), after Cromyosphæra, thus :

E. FIVE SPHERES OR MORE.

Two inner medullary, three outer cortical..............................Caryosphæra.

Ellipsostulus should be inserted (on p. 227, same month and year,) between Ellipsoxiphus and Lithomespilus, thus :—

Two polar spines of different shape.......................................Ellipsostylus.

The key to Druppula I give along with a new key to the whole family of Druppulida as I find the classification given before was not correct. (See p. 228, same month and year, and make the correction).

7. FAMILY, DRUPPULIDA, TWO SHELLS.

A. No solid spines at poles.

Medullary shell simple ; surface smooth..Druppula.
Medullary shell double ; surface spiny.......................................Prunocarpus.

B. Two opposite solid spines at poles.

Medullary shell simple ; spines equal......................................Lithatractus.
Medullary shell simple ; spines unequal...............................Druppatractus.
Medullary shell double ; spines equal...................................... Stylatractus.
Medullary shell double ; spines unequal.................................Xiphatractus.

To be continued.

Mr. Cunningham's Method of Illumination.

BY DR. EDWARD GRAY,

SAN FRANCISCO, CAL.

Mr. Cunningham's article, in the August issue of the JOURNAL proposes the thesis that the diatom is a Protozoan, an animal therefore, and as proof of the assertion he supplies a method of demonstration. Of this he says, at page 231, "an easy method of verification [*i. e.*, of the animal nature of the living diatom] is accessible to all who use the microscope as an instrument of research or for biological studies of any kind." The present article proposes to deal not with the main proposition, but with the kind of evidence upon which it rests. So radical a proposition as is advanced by the Mobile diatomist demands unimpeaceable evidence of its truth. Is Mr. Cunningham's evidence of such character? Let us see. His method of illumination supplying the demonstration is thus described (page 234 :) "In regard to the lighting, and some other requisites of illumination, an Argand, burner lamp is used, a bull's-eye condenser being adjusted as near as possible to the flame, and a large image of the flame projected so as to fall upon the concave face of the mirror. To the sub-stage an achromatic condenser is adapted, and when the light is properly centered in the field, the result will be a dazzling light. "But in order to guarrantee the successful view of the various phenomena, it is necessary to have at hand a glass slip, or a smaller piece of emerald or grass-green colored glass (blue will not answer). This slip must be placed on top of the condenser or the slide containing the living diatoms must rest directly upon the green glass slip."

It is surely a matter of regret that Mr. Cunningham should have left unexplained why blue glass will not answer, while green glass meets the conditions. The wave

lengths obtained by the use of the ammonio-sulphate of copper solution are recognized as supplying the purest mono-chromatic illumination so called. But the gist of the matter lies in the remaining arrangements. A circular flame, with a convex surface therefore, is used in place of a flat flame, then the bull's eye lens is put as close as possible to the flame, whereas it should be at its focal length, the beam now of other than parallel rays is received on the *concave* mirror and sent through the achromatic condenser in what kind of way ? Finally to add to the complexity of matters, a narrow angled half-inch objective is inverted and used as an eye-piece. What the resultant image is would be hard to say ; but observations based on such violations of optical principles governing the obtaining of critical images, can surely not be received as sound. As personal bias cannot, for a moment come into consideration, Mr. Cunningham will doubtless be glad to refresh his memory with the following quotation from Dallinger's revision of Carpenter, p 357 : But the supreme folly of using a concave mirror and a bull's eye is shown in figure 310". . . . this secures a result—as will be seen by the relation of the light to the condenser which is as far from what is sought and desirable as it can well be. While another lesson of great importance may be learnt from fig. 311, which illustrates the error of not having the edge of the flame (e) in the principal focus of the bull's eye (b)."

"The above are fundamental principles of illumination, and if the student is to succeed as a manipulator he must demonstrate and redemonstrate them." Farther, p 362 : "Mr. Nelson has been able to obtain the most wonderful results with narrow cones, 'true ghosts' and 'false ghosts'. . . .and many complex and false images with *the coarser diatoms.* But with wide cones he has proved that these false images *cannot* be produced."

We conclude, then, that Mr. Cunningham's asserted

demonstration fails to demonstrate because of inherent defects. If the diatom is a protozoan that fact has yet to be determined.

EDITORIAL.

A Rare and Valuable Work.–In 1870 and 1871, during the time Dr. Mathias Cook served as a surgeon in the Franco-Prussian war, he came across some parts of an interesting work. Iablonsky's natural system of all domestic and foreign butterflies, as also of the same author domestic and foreign Coleopteras. This work was published by Joachim Pauli, in Berlin, 1783 to 1806.

At that time the doctor's hobby was entomology and he made great efforts to obtain the same. Parts of it were found in Cologne, Bonn, Leipsig, Manheim, Berlin, etc., and in spite of all efforts it was impossible for him to obtain the whole of it. Only one firm in Nurenberg offered, for the sum of $1,000, to get it together, which was rather a trifle more than is generally paid for a book. But nearly 25 years' hard work succeeded in accomplishing a purpose, and with the addition of Mrs. Oberlin's help, who has been fifteen months in Europe, the work was finally obtained from various parties. That part of the work which treats of the day butterflies consists of eleven volumes of reading matter and 327 beautifully colored copper plates, with over 2,200 illustretions. The Coleoptera part consists of ten volumes with 202 colored copper plates, in which are 3,250 illustrations. The most interesting part of the above work is that these 5,000 illustrations are all painted by hand, and so natural that no other process can produce the like. One hundied and eleven years ago, when the work was publised, only 100 copies of the same were issued. Of them only three copies are yet in existence, of which the doctor has one, and it can be safely said that this is the only work of its kind in America.

Tuberculosis in Snakes.—Sometime since a snake was found in England which Mr Arthur Stradling, F. Z. S., of Hertforshire, exhibited before the Natural History Society and which he stated was suffering from consumption or something very analogous to that malady.

MICROSCOPICAL MANIPULATION.

The Staining and Mounting of Tube Casts and other Organic Urinary Deposits.–Bramwell (*British Medical Journal* No. 1, 749, p. 9.) makes the following useful suggestions for the study of urinary sediments. An ordinary conical urine-glass is filled with equal parts of urine and an aqueous solution of boric acid, and set aside until the deposit settles. This is then removed by means of a pipette and transferred to an ordinary test tube containing about half a drachm of a solution of picrocarmin, and the two are thoroughly mixed and set aside for 24 hours. Some of the sediment is then removed by means of a fine mouthed pipette, and mounted. If there is reason to suspect the existence of amylcid disease of the kidney, a solution methyl-violet may be used instead of that of picrocarmin. In order to bring out the fine details of the tube-casts stained in manner described, and in order to preserve them as permanent preparation, they may be mounted in Farrant's solution, consisting of gum arabic and distilled water, each four parts, and glycerin, two parts, with a little camphor. A small test-tube is filled three-quarters with this solution and in it is placed, by means of a fine mouthed pipette; the stained deposit from the test-tube containing the mixture of urine and solution of picrocarmin. The smaller tube is securely corked, inverted two or three times in order to facilitate thorough mixture, and put aside until the sediment has time to settle. In the course of three or four days a minute drop of the deposit is removed from the bottom of the tube by means of a fine mouthed pipette and placed upon a slide and covered. The preparation may, in the course of a few days, be sealed in the ordinary manner. If the prepartion thus mounted is overstained with the solution of picrocarmin, the deposit should be transferred to fresh Farrant's solution. Any organic urinary deposits may, of course, be stained, mounted, and preserved in the same manner.

BACTERIOLOGY.

The Bacillus of the Chinese Bubonic Plague.—The *Lancet* for Aug. 25, contains an article on this subject by Professor S. Kitasato, who gives an interesting account of this dis-

ease and its bacteriological character. The bacilli, he says, are to be found in the blood, in the buboes, in the spleen, and in all other internal organs of the victims of the plague. The bacilli are rods with rounded ends, which are readily stained by the ordinary aniline dye, the poles being stained darker than the middle part, especially in blood preparations, and presenting a capsule sometimes well marked, sometimes indistinct. The bacilli found in the spleen are best stained by a solution of methyl blue. The bacilli show very little movement, and those grown in the incubator, in beef tea, make the medicine somewhat cloudy. The growth of the bacilli is strongest on blood serum at the normal temperature of the human body. Under these conditions they develop luxuriantly, and are moist in consistence and of a yellowish-gray color. They do not liquefy the serum. On agar-agar jelly they also grow freely. The different colonies are of a whitish-gray color, and by a reflected light have a bluish appearance; under the microscope they appear moist and in rounded patches with uneven edges.

At first they appear everywhere as if piled up with "glass-wool," and later as if having dense large centers. If a cover-glass preparation is made from a cultivation on agar-agar, and after having been stained is observed under the microscope, long threads of bacilli are seen which might on careless inspection, be mistaken for a coccus chain, but are recognized with certainty as threads of bacilli under closer observation. The growth on agar-gelatin is similar to that on agar-agar. In a puncture cultivation at the ordinary temperature after a few days they are found growing as a fine dust in little points alongside the puncture, but with very little growth on the surface.

Dr. Kitasato experimented on animals and found that mice, rats, guinea-pigs and rabbits were susceptible to inoculation. If they are inoculated with pure cultivations, or with the blood of a patient in which the bacilli have been observed, or with the contents of a bubo, or with pieces of the internal organs, or even with the contents of the intestine, they become ill in from one to two days, according to the size of the animal. Their eyes become watery, they show disinclination for any effort; later they avoid their food, and hide quietly in a corner of the cage. The parts around the point of inoculation are infiltrated

with a reddish, gelatinous exudation, the spleen is enlarged; sometimes there is swelling of the lymphatic glands, and bacilli are found in all the organs. The results found after death in animals are very similar to those found in anthrax and in Œdema malignum. Pigeons do not appear to be susceptible to the influence of the bacilli.

Trichinosis.—At a recent meeting of the Buffalo Academy of Medicine Frank J. Thornbury, M. D., Buffalo, N. Y., made a preliminary report on five hundred cases of trichinosis in swine observed in his work as Inspector in the Bureau of Animal Industry of the U. S. Department of Agriculture. Special attention was given to conditions in the pathology of trichinosis not previously noted, comprising peculiarities of encapsulation, degenerations, calcifications, pigmentations, etc. A large number of photographs and drawings were presented which illustrated these conditions, and an extensive exhibit of rare and interesting slides under the microscope was given.

Reference was made to the relative frequency and extent of infection of the different parts examined according to the Governement system of inspection. These comprise (1) the diaphram, (2) neck, and (3) loin respectively. The entire number of cases in which trichinæ were found in the diaphram were 400, in the loin 290, and in the neck 170. The average number of trichinæ found in the diaphram in the entire number of cases was 8, in the loin 5, and in the neck 3. In the 500 cases studied all of the three parts were infected in 200 instances, two parts were infected in 136, and one part only was infected in 164 cases. The point of predilection for the trichinæ, therefore, appears to be the diaphram. This is explained by its close proximity to the digestive tract, from which the trichinæ primarly bore. It would appear, also, that the tenderloin is not a preferred article of diet, this comprising as it does the psoas muscles. Where the hogs were extensively infected the trichinæ were also found in the hams, shoulders, sides, and, in fact, in almost every part of the carcass. He has also studied the subject of trichinosis in the human, with the following results: Of twenty-one subjects examined in the dissecting-room in the University of Buffalo, trichinæ were found in the muscles of three. The muscles principally affected were

those of the extremities (one slice from the biceps of an arm containing fifty of the parasites), the diaphram, intercostals, abdominal muscles, psosas, etc. Many of the trichinæ were old and calcified, others were still alive. One of the infections was not very extensive, trichinæ being scattered in limited numbers through the muscles of the body.—*Cincinnati Lancet-Clinic.*

MICROSCOPICAL SOCIETIES.

LINCOLN MICROSCOPE CLUB.—*Roscoe Pound, Secretary.*

October 31. Prof. Hyde of the Lincoln Normal was elected to membership. Dr. Bessey exhibited a small microtome made by Queen and Co. Also a large reading glass arranged for use as a dissecting lens. It can be used as a binocular and enables one to dissect without bringing the eyes close to the lens.

Mr. Pound exhibited some slides of fungi not usually met with: Sporoschisma mirabile, Cladotrichum coolsei, and Olpitrichum polycladum.

There will be no meeting in December on account of the annual meeting of the Nebraska Academy of Sciences.

WASHINGTON, D. C.,

The officers of the Microscopical Society elected for 1894-'95 are as follows:

Prest , W. W. Alleger, M. D.; Vice-Pres., Collins Marshall, M. D.; Cor. Sec., F. E. Maxcy, M. D.; Rec. Sec., Mr. L. M. Mooers; Treas., E. A Balloch, M. D.; Curator, W. H. Seaman, M. D.;

Essay Committee: Dr. E. A. Gibbs; Dr. V. A. Moore; Mr. H. H. Doubleday; and Ex Officio, the coresponding secretary.

Membership committee: Dr. Robt. Reyburn ; Dr. W. H. Seaman; Dr. C. T. Caldwell.

December, 11, 1894.–The regular meeting was held at the rooms of the Society, No. 714 Thirteenth Street N. W. at 8 o'clock Program; A method of conserving micro-anatomical specimens. Projection slides for illustrating lectures, lantern exhibit, by J. Melvin Lamb, M. D.

NEW PUBLICATIONS.

Laboratory Exercises in Botany Designed for the Use of Colleges and Schools in which Botany is Taught by Laboratory Methods. By Edson S. Bastin. 8° 540 pp. 250 figures. Phila., 1895. $2.50.

We have frequently complained in these columns of the waste of time inflicted upon botanical students because their attention has been taken up in classifying and drying specimens as a sequel to the learning of a new language—the technical terms in which this science has always reveled. The present book meets our demands most fully—that students be taught a great deal about a few plants instead of a smattering regarding all plants. To know all that Professor Bastin teaches regarding the Ox-eye Daisy is better than to know all that Linnæus knew about all plants.

The first half of the book is upon Organography and includes the study of roots, stems, leaves, flowers and fruits somewhat after the style of the older botanies, but with new and improved methods. On the very first page one is introduced to the dissecting microscope. The language is a guide to actual laboratory experiments in lieu of a lot of stuff to memorize and repeat in class.

But the second half of the book is what fills us with enthusiasm. This is entitled Vegetable Histology. In it are studies of parenchyma, collenchyma, epidermal tissue, suberous tissue, wood-cells, tracheary tissues, starches, aleurone-grains, chloroplasts, inulin, vasal, collateral and radial bundles etc. In all cases the reader is supposed to actually perform the manipulations needed to demonstrate the structures. Of course this involves a knowledge of microscopy and this book may fairly be said to include a treatise on that subject. The apparatus is described and its use made plain. The stains, media, etc , are fully set forth.

Hence, although written for a college hand-book, it is of the greatest value to all microscopists studying plant structure. There is no serious criticism to be made on the book and no disappointment in store for the purchaser. The price, too, is adapted to our times. For such a book, it is remarkably low and the book will stand the wear of laboratory handling.

THE AMERICAN

MONTHLY MICROSCOPICAL JOURNAL.

INDEX.

LIST OF ILLUSTRATIONS.